PB86-118312

Drinking Water Criteria Document for Polychlorinated Biphenyls (PCBs)

Environmental Criteria and Assessment Office
Cincinnati, OH

April 1985

U.S. Department of Commerce
National Technical Information Service
NTIS

United States
Environmental Protection
Agency

FINAL DRAFT
ECAO-CIN-414
April, 1985
EPA-600/X-84-198-

PB86-118312

EPA

Research and Development

DRINKING WATER CRITERIA DOCUMENT FOR POLYCHLORINATED BIPHENYLS (PCBs)

Prepared for

OFFICE OF DRINKING WATER

Prepared by

Environmental Criteria and
Assessment Office
Cincinnati OH 45268

DRAFT: DO NOT CITE OR QUOTE

NOTICE

This document is a preliminary draft. It has not been formally released by the U.S. Environmental Protection Agency and should not at this stage be construed to represent Agency policy. It is being circulated for comments on its technical accuracy and policy implications.

REPRODUCED BY
NATIONAL TECHNICAL
INFORMATION SERVICE
U.S. DEPARTMENT OF COMMERCE
SPRINGFIELD, VA. 22161

DISCLAIMER

Mention of trade names or commercial products does not constitute endorsement or recommendation for use.

AVAILABILITY NOTICE

This draft document is for internal review and should not be released. For information contact Dr. Jerry F. Stara, Director, Environmental Criteria and Assessment Office, Cincinnati, OH (513/684-7531).

TECHNICAL REPORT DATA

(Please read Instructions on the reverse before completing)

2.

3. RECIPIENT'S ACCESSION NO.
PB86-118312/AS

SUBTITLE
Drinking Water Criteria Document for
POLYCHLORINATED BIPHENYLS (PCBs)

5. REPORT DATE
4/85

6. PERFORMING ORGANIZATION CODE

OR(S)
Environmental Protection Agency

8. PERFORMING ORGANIZATION REPORT NO.

PERFORMING ORGANIZATION NAME AND ADDRESS
Same as box 12.

10. PROGRAM ELEMENT NO.

11. CONTRACT/GRANT NO.

12. SPONSORING AGENCY NAME AND ADDRESS
U.S. Environmental Protection Agency
Office of Drinking Water (WH-550)
401 M St., S.W., Washington, D.C. 20460

13. TYPE OF REPORT AND PERIOD COVERED
Scientific review

14. SPONSORI GENCY CODE

15. SUPPLEMENTARY NOTES

16. ABSTRACT

The Office of Drinking Water (ODW), U.S. Environmental Protection Agency has prepared a Drinking Water Criteria Document on PCBs.
This Criteria Document is an extensive review of the following topics:

- Physical and chemical properties of PCBs
- Toxicokinetics and human exposure to PCBs
- Health Effects of PCBs in humans and animals
- Mechanisms of toxicity of PCBs
- Quantification of toxicological effects of PCBs

17. KEY WORDS AND DOCUMENT ANALYSIS

a. DESCRIPTORS	b. IDENTIFIERS/OPEN ENDED TERMS	c. COSATI Field/Group
PCBs properties PCBs exposure PCBs toxicity Drinking Water		

18. DISTRIBUTION STATEMENT OPEN	19. SECURITY CLASS *(This Report)* Non-sensitive	21. NO. OF PAGES 338
	20. SECURITY CLASS *(This page)* Non-sensitive	22. PRICE

EPA Form 2220-1 (Rev. 4-77) PREVIOUS EDITION IS OBSOLETE

ii

DOCUMENT DEVELOPMENT

Michael L. Dourson, Ph.D., Document Manager
Environmental Criteria and Assessment Office, Cincinnati
U.S. Environmental Protection Agency

Helen H. Ball, M.S., Project Officer
Environmental Criteria and Assessment Office, Cincinnati
U.S. Environmental Protection Agency

Authors

Paul Goetchius, D.V.M.
D. Anthony Gray, Ph.D.
Marijean B. Remington, B.S.
Denzini L. Tullis, Ph.D.
Michael W. Neal, Ph.D.
Syracuse Research Corporation
Syracuse, New York

Muriel Lippman, Ph.D.
Consultant to the Carcinogen
Assessment Group
U.S. Environmental Protection Agency

Scientific Reviewers

Krishan Khanna, Ph.D.
Office of Drinking Water
U.S. Environmental Protection Agency
Washington, DC

Herbert Cornish, Ph.D.
University of Michigan, Retired
Ann Arbor, Michigan

Marion Ehrich, Ph.D.
ICAIR
Life Systems, Inc.
Cleveland, OH

Scientific Reviewers (cont.)

Jennifer Orme, M.S.
Environmental Criteria and Assessment Office, Cincinnati
U.S. Environmental Protection Agency

Steven H. Safe, Ph.D.
ICAIR
Life Systems, Inc.
Cleveland, OH

Ellen Silbergeld, Ph.D.
22 East Mt. Vernon Place
Baltimore, MD

Editorial Reviewers

Erma Durden, B.S.
Environmental Criteria and Assessment Office, Cincinnati
U.S. Environmental Protection Agency

Judith Olsen, B.A.
Environmental Criteria and Assessment Office, Cincinnati
U.S. Environmental Protection Agency

Document Preparation

Technical Support Services Staff: C. Cooper, P. Daunt, C. Fessler, K. Mann, B. Zwayer, Environmental Criteria and Assessment Office, Cincinnati

TABLE OF CONTENTS

Page

I. SUMMARY I-1

II. PHYSICAL AND CHEMICAL PROPERTIES. II-1

STRUCTURE AND IDENTIFICATION. II-1
PHYSICAL AND CHEMICAL PROPERTIES. II-3
STABILITY IN WATER. II-3
SUMMARY II-12

III. TOXICOKINETICS. III-1

ABSORPTION. III-1

Gastrointestinal Absorption. III-1
Dermal Absorption. III-3
Inhalation III-4

DISTRIBUTION. III-5

Studies in Rats. III-5
Studies in Mice. III-8
Studies in Primates. III-8
Studies on PCB Mixtures to Rodents III-10
Distribution to the Fetus and Neonate (Rodents). III-12
Distribution to the Fetus and Neonate (Primates) III-18

BIOACCUMULATION AND RETENTION III-21
METABOLISM. III-23

Metabolism of Mono-, Di- and Tri-chlorinated Isomers III-24
Metabolism of Penta- and Hexa-chlorinated Isomers. III-29

EXCRETION III-32

Rodent Studies III-32
Studies in Monkey and Man. III-38
Alteration of Excretion. III-39

SUMMARY III-43

IV. HUMAN EXPOSURE. IV-1

EXPOSURE ESTIMATION IV-1

Drinking Water IV-1
Diet IV-3
Air. IV-3

SUMMARY IV-5
REFERENCES. IV-7

Page

V. HEALTH EFFECTS IN ANIMALS V-1

GENERAL TOXICITY. V-1

Acute and Short-Term Toxicity. V-1
Chronic and Subchronic Toxicity. V-26

TARGET ORGAN TOXICITY V-44

Hepatic Effects. V-44
Immunological Effects. V-52
Thyroid Effects. V-55

CARCINOGENIC, MUTAGENIC AND REPRODUCTIVE EFFECTS. V-59

Carcinogenicity. V-59
Mutagenicity V-94
Teratogenicity and Reproductive Toxicity V-103

SUMMARY V-125

VI. HEALTH EFFECTS IN HUMANS. VI-1

CASE STUDIES. VI-1

Occupational VI-1
Clinical Case Studies. VI-13

EPIDEMIOLOGICAL STUDIES VI-14

Yusho Incident VI-14
Yu-Cheng Incident. VI-21
Effects on Blood Pressure Measurements VI-23
Ocular Melanoma. VI-23

HIGH RISK SUBPOPULATIONS. VI-24
SUMMARY VI-26

VII. MECHANISMS OF TOXICITY. VII-1

ENZYME INDUCTION. VII-1
INHIBITION OF MITOCHONDRIAL RESPIRATION VII-2
REPRODUCTIVE TOXICITY VII-3
MOBILIZATION AND REDISTRIBUTION OF STORED PCBs. VII-4
COVALENT BINDING. VII-5
ALTERED CALCIUM METABOLISM. VII-6
IMMUNOLOGICAL EFFECTS VII-6
THYROID TOXICITY. VII-7
METABOLISM. VII-7
INTERACTIVE EFFECTS VII-9
SUMMARY VII-11

Page

VIII. QUANTIFICATION OF TOXICOLOGIC EFFECTS VIII-1

EXISTING GUIDELINES, RECOMMENDATIONS AND STANDARDS. VIII-4
NONCARCINOGENIC EFFECTS VIII-5
QUANTIFICATION OF NONCARCINOGENIC RISK. VIII-14
CARCINOGENIC EFFECT VIII-16
QUANTIFICATION OF CARCINOGENIC RISK VIII-19
SPECIAL CONSIDERATION VIII-21

High Risk Subpopulation. VIII-21
Co-Carcinogenesis, Initiation and Promotion. VIII-21

SUMMARY VIII-22

IX. REFERENCES. IX-1

LIST OF TABLES

No.	Title	Page
II-1	CAS and RTECS Numbers for PCBs.	II-2
II-2	Properties of Aroclors.	II-4
II-3	Percent Composition of Aroclors and Kanechlors.	II-6
II-4	Polychlorinated Dibenzofuran Concentration in Commercial PCBs	II-7
III-1	Uptake of PCBs by Rats.	III-2
III-2	Tissue Distributions of Pure PCB Isomers Given to Animals .	III-6
III-3	Retention of PCBs in Mouse Fat at 7 and 21 Days After Dosing.	III-9
III-4	Tissue Distributions of PCB Mixtures Given to Animals . . .	III-11
III-5	Distribution of PCBs to Fetuses and Nursing Young	III-13
III-6	Half-lives of PCBs.	III-34
III-7	Comparative Rates of Elimination of Some Major PCBs from the Blood of PCB-Poisoned Patients.	III-40
IV-1	Estimated Dietary Intake of PCBs for Adults, Infants and Toddlers.	IV-4
IV-2	Estimated Intake of PCBs from the Environment by Adult Males	IV-6
V-1	Lethal Doses of PCBs.	V-2
V-2	Acute and Short-Term Toxicity of Orally-Administered PCBs to Rats.	V-5
V-3	Acute and Short-Term Toxicity of Orally-Administered PCBs to Mice.	V-9
V-4	Acute and Short-Term Toxicity of PCBs Administered by Routes Other Than Oral	V-13
V-5	Acute and Short-Term Toxicity of Orally-Administrated PCBs to Other Species	V-17
V-6	Effects of Subchronic and Chronic Oral Exposure of PCBs to Rats	V-27

No.	Title	Page
V-7	Subchronic and Chronic Oral Exposure of PCBs to Mice. . . .	V-29
V-8	Effects of Chronic and Subchronic Exposure of PCBs to Monkeys	V-30
V-9	Effects of Subchronic and Chronic Oral Exposure of PCBs to Other Species.	V-33
V-10	Weight Data; Further Analysis of Changes.	V-64
V-11	Percentage Average Weight Increased Calculated.	V-66
V-12	Proliferative Lesions of the Liver in Fischer Rats Fed Aroclor 1254.	V-68
V-13	Effects on Liver Tumorigenesis and Carcinogenesis: Feeding Studies Using Various Commercial PCB Preparations .	V-74
V-14	Number of Isomers at Each Chlorinated Level for Three Different ARoclor 1260 Preparations	V-77
V-15	Comparisons of Hexachlorobiphenyl Isomers of Aroclor 1260 Showing the Qualitative and Quantitative Variation in Three Different Aroclor Preparations	V-78
V-16	Analyses of Three Different Aroclor 1260 and Three Different Aroclor 1254 Preparations for the Presence of PCB Isomers Known to Utilize, to Some Degree, A Metabolic Pathway which Forms Arene Oxide Intermediates	V-81
V-17	Action of Kanechlor 500 and α-, β- or γ-Benzene Hexachloride.	V-86
V-18	Percent Weight Change in Kanechlor 500 Exposed Rats	V-89
V-19	The Liver and Body Weight Ratio and the Number of Liver Tumors Produced in Offspring Rats Exposed to PCB through Their Dams and Treated with DEN after Weaning	V-91
V-20	Summary of Mutagenicity Assays of PCBs in Salmonella typhimurium	V-95
V-21	Summary: Teratogenetic, Fetotoxic and Reproductive Effects of Orally Administered PCBs	V-104
VI-1	Summary of the Symptoms in Sixteen Cases of Acneform Eruption.	VI-2
VI-2	Duration of PCB Exposure of 326 Capacitor Manufacturing Workers	VI-5

No.	Title	Page
VI-3	Age Distribution of 326 Capacitor Manufacturing Workers by Sex.	VI-6
VI-4	Prevalence of Reported Dermatologic Symptoms Among 326 Capacitor Manufacturing Workers	VI-8
VI-5	Prevalence of Gastrointestinal, Musculoskeletal and Neurologic Symptoms by Symptom Category and Sex Among 326 Capacitor Manufacturing Workers	VI-9
VI-6	Clinical Features of Six Electrical Workers with Chloracne.	VI-11
VI-7	Initial Symptoms of Yusho Among 136 Persons	VI-15
VI-8	Percent Distribution of Symptoms of Yusho Among 189 Persons	VI-16
VI-9	Relationship Between the Amount of Kanechlor-Contaminated Rice Oil Consumed and Clinical Severity of Yusho.	VI-18
VI-10	Relationship Between Clinical Severity of Yusho and Age of Affected Persons	VI-19
VI-11	Concentration of PCBs and PCDFs in the Toxic Rice-Bran Oils That Caused "PCB Poisoning" in Taichung, Taiwan.	VI-22
VIII-1	FDA Regulations for PCBs.	VIII-6
VIII-2	Data Used as the Basis for the q_1^*.	VIII-20
VIII-3	Summary of Health Advisories and Lifetime Cancer Risk for PCBs	VIII-23

LIST OF ABBREVIATIONS

AADI	Adjusted acceptable daily intake
AAH	Aryl hydrocarbon hydroxylase
ADI	Acceptable daily intake
BCF	Bioconcentration factor
BHC	Benzene hexachloride
BOD	Biologic oxygen demand
bw	Body weight
CB	Chlorinated biphenyl
DCB	Dichlorinated biphenyl
DEN	Diethylnitrosamine
DMBA	Dimethylbenzanthracene
DMN	Dimethylnitrosamine
DNA	Deoxyribonucleic acid
EXAMS	Exposure Assessment Modeling System
2-FAA	N-2-fluoroenylacetamide
GI	Gastrointestinal
gmw	Gram molecular weight
HA	Health Advisory
HCB	Hexachlorinate biphenyl
i.p.	Intraperitoneal
LD_{50}	Lethal dose for 50% of recipients
LOAEL	Lowest-observed-adverse-effect level
LOEL	Lowest-observed-effect level
3-MC	3-methylcholanthrene
MCB	Monochlorinated biphenyl
3'-MeDAB	3'-methyl-4-dimethylaminoazobenzene
MFO	Mixed function oxidase
MNNG	N-methyl-N'-nitro'N'-nitrosoquanidine
NOAEL	No-observed-adverse-effect level
PCB	Polychlorinated biphenyl
PCDF	Polychlorinated dibenzofuran
PCE	Polychromatic erythrocytes
PeCB	Pentachlorinated biphenyl
RNA	Ribonucleic acid
s.c.	Subcutaneous
SNARL	Suggested no adverse response level
STEL	Short-term exposure limit
TCB	Tetrachlorinated biphenyl
TEM	Triethylenemelamine
TPA	12-O-tetradecanoylphorbol-13-acetate
triCB	Trichlorinated biphenyl
TWA	Time weighted average
UV	Ultraviolet

I. SUMMARY

The evaluation of the health effects of polychlorinated biphenyls (PCBs) in the environment represents a highly complex problem. PCB commercial formulations enter the environment as mixtures, which represent a variety of individual isomers, congeners and impurities. Which of the components of the chemical mixtures is responsible for the adverse health effects is not known. However, individual isomers have been tested for some of the adverse health effects.

PCBs, formerly marketed principally by Monsanto under the trade name Aroclor, are a family of compounds commercially available as mixtures. The physical properties of the component isomers vary considerably, with a molecular mass range of 154-499, a log octanol/water partition coefficient range of 3.79-11.20, and a solubility range of 9.77×10^{-10} to 4.68×10^{-5} moles/ℓ (moles/ℓ = gmw x 10^{-6}/ℓ = μg/ℓ).

PCBs will volatilize from ambient waters, with half-lives varying from 2 months to >150 years, as predicted by an EXAMS model. The most sensitive input parameter affecting volatilization rates was found to be the octanol/water partition coefficient (and those partition coefficients calculated from it, e.g., sediment, organic content, biota), indicating that the partitioning of the PCBs from the sediments and biota into the water was the limiting factor affecting volatilization. Adsorption appears to be the dominant removal mechanism from water for chlorinated PCBs. Based on information obtained from an EXAMS model, adsorption to sediment will effectively bind between 45 and >99% of the PCBs present in water depending on the PCB and the organic matter present in the sediment. The higher the organic matter in the sediment or the higher the chlorination, the more strongly adsorbed is the PCB mixture or isomer.

As with volatilization, biodegradation is significant for only the less chlorinated species. PCBs containing three or fewer chlorines will be degraded in the environment; estimating the half-lives is difficult, however, because of variability in the reported literature. PCBs with four chlorines appear to be only somewhat degradable, while those with five or more appear to be very slowly degraded, if at all (Brown et al., 1984).

Absorption of ingested PCBs from the gastrointestinal tract of animals tends to be almost complete, usually >90%. Absorption through the skin may also be high, judging from the reports of mortality and other signs of toxicity, although quantitative data were not found.

Distribution to body tissues appears to be biphasic. An initial distribution to muscle and liver occurs quite rapidly and blood levels fall shortly thereafter; over time, liver and muscle concentrations fall and concentrations in adipose tissue, skin and lipid-containing organs increase.

Transplacental movement of PCBs occurs, with PCB levels in fetal tissue slightly lower than those of dams. During the later stages of gestation, transfer across the placenta seems to increase, probably because of fetal ingestion of amniotic fluid. In final gestation stages, PCB levels in fetal tissues may exceed the levels in dams. Postnatally, PCBs are transferred via lactation resulting in rapidly elevated PCB levels in tissues of offspring. Concentrations of PCBs in milk exceed those of maternal blood;

as a result, concentrations of PCBs in tissues of offspring may be 5-10 times those in the tissues of dams. In general, those isomers that are stored longest at higher concentrations in body fat are the more highly chlorinated PCBs. They are also the isomers that are transferred in higher concentrations in milk. Breast milk of nursing women has been found to contain significant levels of PCBs.

Metabolism of the less chlorinated isomers generally occurs more rapidly than does metabolism of more highly chlorinated isomers. Additionally, the position of chlorine substituents substantially influences the rate and extent of PCB metabolism.

Metabolism in animals results in the formation of mono- and dihydroxy-PCBs, which are conjugated when excreted in the urine and may or may not be conjugated if excreted from the bile. Additionally, sulfoxy-, methyl- and methoxy- groups may be attached to derivatives of PCBs. In rabbits and monkeys, trans-dihydrodiols are formed as a predominant metabolite, which strongly suggests metabolism through an arene oxide intermediate. Arene oxides have been implicated in cellular necrosis, mutagenicity and carcinogenicity.

Fecal excretion of unchanged PCB isomers may account for as much as 23% of the total excretion. Apparently, unchanged PCB isomers are secreted directly into the lumen of the duodenum; however, most of the excretion occurs through the biliary route. Urinary excretion accounts for up to 12% of the excretion of most isomers and as much as 50% of the excretion of a monochlorinated isomer.

Phenobarbital and phenobarbital-type inducers increase the rate of metabolism of PCBs, and hence their elimination. Weight loss and mobilization of fat stores increases blood levels and excretion of PCBs. Activated charcoal reduces G.I. absorption of PCBs.

Most PCB mixtures exhibit rather low oral toxicity relative to other polychlorinated hydrocarbons. Reported LD_{50}s range from 0.5 mg/kg bw for 3,4,5,-3',4',5' HBC in guinea pigs upto 10 g/kg bw in rats for commercial mixtures.

Subchronic and chronic exposures to PCBs result in reductions of body temperature as well as reductions in food and water intake at doses as low as 0.46 mg/kg bw in mice. Reduced body weight gain or weight loss often occurs. Necropsy often reveals emaciation and an absence of body fat. Hematocrit and hemoglobin are generally normal; however, a few reports indicate a slight decrease in hemoglobin and hematocrit and a lymphocytopoenia with a compensatory neutrophilia. Also, analysis of blood chemistry has often indicated elevated serum cholesterol, lipids and liver enzymes.

A major target organ for PCBs is the liver, where ~4-fold increases in size have been reported for rats at high-dose levels of 1000 ppm in the diet. This is caused by hepatocellular enlargement that may be due to fatty infiltration and proliferation of smooth endoplasmic reticulum. At high-dose levels, focal necrosis and degeneration occur, iron-containing deposits are found in the Kupfer cells and there is increased urinary excretion of certain porphyrins and precursors. The major effect on the liver seems to be a several-fold increase in the induction of certain hepatic enzymes, particularly the microsomal mixed function oxidases.

PCBs act directly on the thyroid gland to interfere with thyroxin and tri-iodothyronine production. This may be due to the interference with the mechanism by which cleavage of the iodine-rich colloid results in secretion of the two thyroactive hormones. Increased conjugation by the liver may also contribute to reduced serum thyroxin levels. Some of the toxic effects of PCBs (such as dermal hyperpigmentation, weight loss, depressed appetite) have been attributed to thyrotoxic effects.

PCBs also have dramatic effects on the immune system with may be one of the more sensitive indicators of PCBG intoxication. For example, PCBs are associated with thymic atrophy, lymphocytopenia and splenomegaly. Other signs of PCB toxicity in animals include severe facial and subcutaneous edema in monkeys, often with the development of comedones and cysts of the meibomian glands, resembling chloracne in man. Chronic exposure in rats and mice has been reported to cause hair loss and crusty or ulcerated skin lesions.

Monkeys develop gastritis when exposed to PCBs at levels as low as 3 ppm for 35 weeks. Stomach linings become thickened and mucus-containing cysts develop. Parietal and zymogenic cells disappear and the hypertrophic mucosal cells become locally invasive.

Reports of mutagenicity in the Ames assay are conflicting. Most reports indicate a lack of mutagenicity, but one report claims that 4-monochlorobiphenyl causes a marked increase of mutations in the Ames systems. No reports of cytogenetic changes or dominant lethal effects attributable to PCBs have been located in the available literature.

Two authors reported a slight increases in incidence of cleft palate in fetal mice from dams exposed to PCBs during gestation. No other teratogenic effects were found. Several laboratories have reported fetal toxicity that was attributed to PCBs (resorptions, abortions, reduced birth weight and decreased postnatal survival) in several species. Nursing offspring of PCB-exposed dams suffer mortality or other signs of intoxication probably as a result of PCBs being transmitted through maternal milk fat.

The carcinogenic and tumorigenic potential of PCBs has been demonstrated for three PCB compounds. Two PCB compounds have produced hepatocellular carcinomas, Aroclor 1260 in rats and Kaneclor 500 in mice. Further support comes from a rat study using Aroclor 1254. While the results were not statistically significant, the elevated response does indicate the tumorigenic potential of Arachlor 1254. PCBs may also inhibit the growth of transplantable tumors, although the mechanism of this effect is currently unknown.

Both occupational exposure to dermal contact and airborne concentrations of PCBs and accidental ingestion of rice oil contaminated with PCBs induced similar dermatologic and hepatic effects in humans. The available epidemiology data for cancer does not provide sufficient information to conclude whether or not PCB exposure results in increased cancer response. Infants born to women who had Yusho during pregnancy were generally small and cola-colored, and had other minor defects such as early eruption of teeth and gingival hyperplasia. High risk subpopulations were identified as those with the potential for frequent or high exposures, including occupationally-exposed workers, nursing infants exposed to PCBs from human breast milk and those individuals with the inability to excrete PCBs via glucuronidation, thereby decreasing their capability to detoxify and excrete these xenobiotic compounds.

Several mechanisms have been proposed to explain the toxicity of PCBs. These include enzyme induction and inhibition of mitochondrial respiration. Enzyme induction appears to be of either the 3-methylcholanthrene (P-448) and/or phenobarbital (P-450) type, depending on positions of chlorination. Long-term toxicity seems to be related to 3-methylcholanthrene-type induction. Reproductive toxicity appears to be associated with a direct gonadotoxic effect exhibited by PCBs or by induced catabolism of steroid hormones.

Metabolism of PCBs may produce an electrophilic epoxide intermediate which can bind covalently to cellular macromolecules such as DNA, RNA or proteins. Mutagenic effects, carcinogenic effects or cellular necrosis may result.

The ability of PCBs to cause immunosuppression is a very sensitive indicator of toxicity. It is manifested as a reduction in levels of circulating antibodies and as increased mortality after challenge by endotoxin or pathogenic organisms. It may be mediated by suppression of RNA synthesis in macrophages resulting in interference with the ability to acquire and transfer immunity.

PCBs also alter cellular metabolic handling of other compounds, usually by inducing enzyme systems that result in their hastened metabolism. Cadmium exerts an antagonistic effect on many aspects of the toxicity of PCBs, apparently by inhibiting the enzyme systems that PCBs induce.

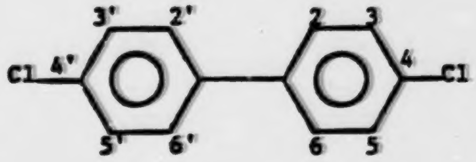
The 1-day HAs for a 10 kg child and a 70 kg adult are 2.9 and 14 μg/ℓ; the 10-day HAs for a child and an adult are 2.5 and 8.8 μg/ℓ, respectively. A lifetime AADI is not recommended. Based on an increased lifetime risk of cancer of 10^{-4}, 10^{-5} and 10^{-6}, the corresponding PCB levels in the drinking water are 0.8, 0.08 and 0.008 μg/ℓ.

II. PHYSICAL AND CHEMICAL PROPERTIES

Structure and Identification

Polychlorinated biphenyls (PCBs) are a family of compounds that vary widely in physical, chemical and biological properties. There are 209 possible compounds resulting from the total or partial chlorination of a biphenyl, of which some 100 individual compounds have been detected (Hutzinger et al., 1974). In the designation of the individual PCBs, a set of four digits was used by Monsanto (the U.S. manufacturer), who refer to their PCBs as Aroclors. The first two digits indicate that the preparation is a mixture (12) and the second two digits indicate the chlorine content percentage by mass. Thus, Aroclor 1242 is a mixture with an average chlorine content of 42%. When it was determined that there were significant environmental problems with heavily chlorinated species, Monsanto developed a mixture that was primarily mono-, di- and trichloro isomers. This was designated Aroclor 1016.

The structure of a typical PCB (4,4'-dichlorobiphenyl) is shown:



The Chemical Abstracts Service (CAS) registry numbers and the Registry of Toxic Effects of Chemical Substances (RTECS) numbers for representative PCBs given in Table II-1.

TABLE II-1

CAS and RTECS Numbers for PCBs*

Aroclor Number	CASS Number	RTECSS Number
1016	12674-11-2	NA
1221	11104-28-2	TQ 1352000
1232	11141-16-5	TQ 1354000
1242	53469-21-9	TQ 1356000
1248	12672-29-6	TQ 1358000
1254	11097-69-1	TQ 1360000
1260	11096-82-5	TQ 1362000

*Source: NIOSH, 1983

NA = Not available

Physical and Chemical Properties

Properties of PCBs are summarized in Table II-2. Chemical formulas and molecular weights of PCBs are listed in Table II-3. It should be noted that the properties of the Aroclors listed are average values, since the Aroclors are mixtures. For example, molecular mass of the PCBs ranges from 154 for biphenyl to 499 for decachlorobiphenyl, log octanol/water partition coefficients range between 3.79 and 11.20, and solubility ranges between 9.77×10^{-10} and 4.68×10^{-5} moles/ℓ (moles/ℓ = gmw x 10^{-6}/ℓ = μg/ℓ) (Yalkowsky et al., 1983).

Bowes et al. (1975) reported finding polychlorinated dibenzofurans (PCDF) in commercial PCBs manufactured in the United States, Germany and France. Table II-4 lists the results for Aroclors 1248, 1254, 1260 and 1016 (United States), Clophen A-60 (German) and Phenoclor DP-6 (French). Levels ranged between 0.8 and 13.6 μg/g, the highest concentrations being in the German and French products. Roach and Pomerantz (1974) and Nagayama et al. (1976) detected PCDFs in Japanese PCBs (Kanechlors). Roach and Pomerantz (1974) estimated -1 ppm of the tetrachlorodibenzofuran was present in the Kanechlor KC-400 sample. Nagayama et al. (1976) reported the following PCDFs in Kanechlors and Yusho oil (rice oil): KC-300k 1-1.5 ppm of tetra- and penta-CDF; KC-400, 17-18 ppm of di-, tri-, tetra- and penta-CDF; KC-500, 2.5-4 ppm of penta-, hexa- and hepta-CDF; KC-600, 3-5 ppm of tetra-, penta-, hexa- and hepta-CDF; Yusho oil, 4-5 ppm of tri-, tetra-, penta- and hexa-CDF. Silbergeld (1983) gives concentrations of chlorinated dibenzofurans in several commercial PCB products. Values of tetra-, penta- and hexa-chlorinated dibenzofurans range from "not detected" in Arochlor 1016 to as high as 13.6 ppm in Phenoclor DP-6.

TABLE II-2

Properties of Aroclors^{a,b,c}

Property	VALUE						
	1016	1221	1232	1242	1248	1254	1260
Appearance	clear oil	clear oil	clear oil	clear oil	clear oil	light yellow viscous liquid	light yellow sticky resin
Chlorine (percent)	41	20.5-21.5	31.4-31.5	42	48	54	60
Density (gm/cm^3) (25°C)	1.33	1.15	1.24	1.35	1.41	1.50	1.58
Distillation range (°C)	325-356	275-320	290-325	325-366	340-375	365-390	385-420
Evaporation loss %100°C/6 hours		1-1.5	1-1.5	0-0.4	0-0.3	0-0.2	0-0.1
Aqueous solubility mg/ℓ	0.42^d	0.59^i	NA	0.24,0.34^d 0.13^c	0.054	0.012, 0.024^e 0.056^j	0.027
Lipid solubility (organic solvents)	very soluble	very soluble	very soluble	very soluble	very soluble	very soluble	very soluble
Vapor pressure (mm Hg at 25°C)	[4x10^{-4}]	[6.7x10^{-3}]	[4.06x10^{-3}]	4.06x10^{-4}	4.94x10^{-4}	7.71x10^{-5}	4.05x10^{-5}
Log octanol/water partition coefficient	4.38^d >5.58^{f,g}	[2.8] 4.09^{g,h}	[3.2] >4.54^{g,h}	4.11^d >5.58^{f,g}	[5.75]^i >6.11^{f,g}	[6.03]^i	[7.14]^i >6.11^{f,g}

TABLE II-2 (cont.)

Property	VALUE						
	1016	1221	1232	1242	1248	1254	1260
Absorption capacity of activated carbon (mg/g)		242[k]	630[k]				
Conversion factors 1 ppm =	10.05 mg/m^3	8.21 mg/m^3	9.50 mg/m^3	10.9 mg/m^3	12.2 mg/m^3	13.4 mg/m^3	15.4 mg/m^3
1 mg/m^3 =	0.0940 ppm	0.122 ppm	0.105 ppm	0.0917 ppm	0.0816 ppm	0.0745 ppm	0.0651 ppm

[a] Adapted from Callahan et al., 1979

[b] All values not subscripted are from Monsanto (1974).

[c] Bracketed data are estimated.

[d] Paris et al., 1978

[e] Dexter and Pavlov, 1978

[f] Chiou et al., 1977

[g] Partition coefficient of lowest chlorinated biphenyl present in significant quantities.

[h] Tulp and Hutzinger, 1978

[i] Harsch et al., 1974; Chiou et al., 1977

[j] Haque et al., 1974

[k] Ramanathan, 1984

[l] Hollifield, 1979

NA = Not available

TABLE II-3

Percent Composition of Aroclors and Kanechlors[a]

Empirical Formula	Aroclor Number							Kanechlor Number		
	1016	1221	1232	1242	1248	1254	1260[b]	300	400	500
$C_{12}H_{10}$	<0.1	11	<0.1	<0.1	ND	<0.1	ND	NR	NR	NR
$C_{12}H_9Cl$	1	51	31	1	ND	<0.1	ND	NR	NR	NR
$C_{12}H_8Cl_2$	20	32	24	16	2	0.5	ND	16.6	3.0	NR
$C_{12}H_7Cl_3$	57	4	28	49	18	1	ND	59.8	32.8	5.0
$C_{12}H_6Cl_4$	21	2	12	25	40	21	1	23.0	43.8	26.5
$C_{12}H_5Cl_5$	1	<0.5	4	8	36	48	12	0.6	15.8	55.0
$C_{12}H_4Cl_6$	<0.1	ND	<0.1	1	4	23	38	NR	4.6	12.8
$C_{12}H_3Cl_7$	ND	ND	ND	<0.1	ND	6	41	NR	NR	NR
$C_{12}H_2Cl_8$	ND	ND	ND	ND	ND	ND	8	NR	NR	NR
$C_{12}HCl_9$	ND	ND	ND	ND	ND	ND	ND	NR	NR	NR
Percent chlorine	41	20.5- 21.5	31.4- 32.5	42	48	54	60	NA	49	54

[a] Source: Callahan et al., 1979; IARC, 1974; Masuda et al., 1974

[b] Kanechlor 600 (Japanese), Phenoclor DP6 (French) and Clophen A60 (German); all contain ~60% Cl (Masuda et al., 1974; Fishbein, 1974).

ND = None detected

NR = Not reported

TABLE II-4

Polychlorinated Dibenzofuran Concentration in Commercial PCBs*

PCB	Concentration (μg/g)			
	4-Cl	5-Cl	6-Cl	Total
Aroclor 1248 (1969)	0.5	1.2	0.3	2.0
Aroclor 1254 (1969)	0.1	0.2	1.4	1.7
Aroclor 1254 (1970)	0.2	0.4	0.9	1.5
Aroclor 1260 (1969)	0.1	0.4	0.5	1.01
Aroclor 1260 (lot AK3)	0.2	0.3	0.3	0.8
Aroclor 1016 (1972)	<0.001	<0.001	<0.001	<0.003
Clophen A-60	1.4	5.0	2.2	8.4
Phenoclor DP-6	0.7	10.0	2.9	13.6

*Source: Adapted from Bowes et al., 1975

Stability in Water

Commercially prepared PCBs are complex mixtures of compounds rather than single components. As such, the fate of the commercial products Aroclors) in the environment is dependent on the components of the mixture. The Aroclors will be discussed primarily because much of the literature is written about them, and their use facilitates the discussion. It should be noted, however, that once in the environment or treatment plant, an Aroclor will change with time because of the selective adsorption, evaporation or biodegradation of specific congeners. The literature concerned with PCBs in water is vast, and this discussion is not intended to be a comprehensive review. The literature presented here, however, appears to be representative of the majority of studies. Based on the studies available, the dominant removal processes for low molecular weight PCBs are biodegradation and volatilization although sedimention also occurs. Higher molecular weight PCBs will principally adsorb to sediments and biota, although some volatilization may occur.

Stability of PCBs in water may also be reduced by photolysis (Pomerantz et al., 1978; Callahan et al., 1979). Although photolytic reactions may occur in the environment, the rate and extent of PCB degradation by photolysis are extremely difficult to assess. Complicating factors include the diversity of environmental conditions and the propensity of PCBs (particularly the more photo-labile highly-chlorinated biphenyls) to adsorb to sediments and organic materials.

Oxidation is not likely to be an important environmental conversion process for PCBs. Although chlorinated dibenzofurans are present in commercial mixtures, their origin is not known, and evidence for their environmental formation is tenuous (chlorinated dibenzofurans are formed in very low yields during oxidation of PCBs) (Pomerantz et al., 1978). PCBs are also unlikely to be affected by hydrolysis as an environmental process because S_N1 and S_N2 reactions do not take place readily at sp^2 hybridized carbons (Morrison and Boyd, 1973).

The occurrence of PCBs in the atmosphere and several laboratory studies has confirmed the importance of volatilization as a removal process for PCBs from water (Doskey and Andren, 1981; Callahan et al., 1979; Atlas et al., 1983; Eisenreich et al., 1981; Mackay and Leinonen, 1975). Volatilization half-life data for environmental waters were not found in the available literature, and modeling the fate of the PCBs was complicated by the numerous reported solubilities, octanol/water partition coefficients, and Henry's law constants for each Aroclor (Callahan et al., 1979; Doskey and Andren, 1981; Mabey et al., 1981). Some Henry's law constants varied over four orders of magnitude for the same Aroclor (Doskey and Andren, 1981), thus no criteria could be developed to choose one value over another. Based on a number of input values for these parameters, however, the volatilization half-lives for various Aroclors calculated by EXAMS (Burns et al., 1981) appear to be in the following ranges: Aroclor 1221, 2 months to 1 year; Aroclor 1232, 2 months to 1 year; Aroclor 1016, <2-7 years; Aroclor 1242, 2-7 years; Aroclor 1248, 3-8 years; Aroclor 1254, >4-11 years; Aroclor 1260, >60 to >150 years.

One of the major controlling factors in determining the volatilization half-life with EXAMS (Burns et al., 1981) was the octanol/water partition coefficient since this determines the amount of PCBs partitioning into the water from sediments and biota, and available for volatilization. Again, several values for the octanol/water partition coefficient were found to vary over two orders of magnitude varying over two orders of magnitude were found for the Aroclors (Callahan et al., 1979; Garten and Trakalka, 1983). The value of using the EXAMS model (Burns et al., 1981) in this situation is that adsorption is taken into account when calculating volatilization half-lives, while models for volatilization alone do not (Mackay and Leinonen, 1975). Hence, the half-lives presented above, though approximate, do have more meaning than the half-lives from other models predicting volatilization alone in the absence of adsorption.

The reduction of volatilization half-lives in the presence of adsorption has been studied somewhat in the laboratory. Oloffs et al. (1972, 1973) showed that Aroclor 1260 (100 μg/ℓ) volatilized 67% from river water after 12 weeks but only 34% volatilized when sediment was added. Tucker et al. (1975) reported that Aroclors 1221 and 1016 volatilized 4.2 and 3.6% from aerated samples containing activated sludge.

Adsorption onto sediments and other organic matter, including treatment plant sludge, coagulant and possibly plastic water pipe and coal tar coated water pipe, is possibly the dominant removal process for high molecular weight PCBs in water. Based on a number of the octanol/water partition coefficients available (and those partition coefficients calculated from them, e.g., sediment, organic content, biota) (Callahan et al., 1979; Mabey et al., 1981; Mackay, 1982), the EXAMS model (Burns et al., 1981) predicted that the following amount of PCB would be adsorbed onto sediments: Aroclor 1221, 45-95%; Aroclor 1232, 70-98%; Aroclor 1016 and 1242, 96->99%; Aroclor 1248, 97->99%; Aroclor 1254, 98->99% and Aroclor 1260, >99%. These values are dependent on the choice of partition coefficient and on the amount of organic matter present in the sediment. These results appear to be in keeping with other experimental data (Oloffs et al., 1973; Haque et al., 1974; Moein et al., 1976; Moein, 1976; Hetling et al., 1978; Paris et al., 1978). The adsorption characteristics of each PCB component of the Aroclors presented will, of course, vary considerably. Thus, the lower chlorinated components will adsorb as strongly to the organic matter than the more highly chlorinated molecules. In addition, the more strongly adsorbed components are also less biodegradable, and their adsorption characteristics make resolution into the water possible for years after they enter sediments, biota and possibly water pipes.

A number of investigators have reported on both the biodegradability and mechanism of biodegradation of PCBs (Hutzinger et al., 1972; Kaiser and Wong, 1974; Branson et al., 1975; Berlin et al., 1975; Wong and Kaiser, 1976; Furukawa et al., 1978, 1983; Tabak et al., 1981a,b). Biodegradability is generally related to the number of hydrogens available. These positions appear to be hydroxylated by microsomal oxidation. If an adjacent position is unchlorinated, degradation is then facilitated by allowing the formation of an arene oxide.

Biodegradation rates vary considerably and are dependent upon many factors including the amount of chlorination, concentration, type of microbial population, available nutrients and temperature. An example of this variation in biodegradation rate was reported by Tucker et al. (1975) for Aroclors in an acclimated, semicontinuous activated sludge experiment with 48-hour exposure: Aroclor 1221, 81±6%; Aroclor 1016, 33±14%; Aroclor 1242, 26±16% and Aroclor 1254, 19±38%. Tabak et al. (1981) found virtually complete degradation for Aroclors 1221 and 1232 at 5 and 10 mg/ℓ concentrations using a static-culture, flask-screening procedure employing BOD dilution water and a settled domestic wastewater inoculum over 28 days. By contrast, Aroclors 1016 and 1242 showed moderate degradation (~40%), while Aroclors 1248, 1254 and 1260 showed almost no degradation.

This type of degradation pattern has been reported by Oloffs et al. (1972), Moein et al. (1976) and Wong and Kaiser (1976). The results show that mono-, di- and trichlorinated biphenyls (Aroclors 1221 and 1232) biodegrade relatively rapidly, tetrachlorinated biphenyls (Aroclors 1016 and 1242) biodegrade slowly and higher chlorinated biphenyls (Aroclors 1248, 1254 and 1260) are recalcitrant.

Summary

Polychlorinated biphenyls (PCBs), marketed in the U.S. principally by Monsanto under the trade name Aroclor, are a family of compounds that were available as mixtures. The physical properties of the components vary considerably with a molecular mass range of 154-499, a log octanol/water partition coefficient range of 2.79-11.20, and a solubility range of 9.77×10^{-10} to 4.68×10^{-5} moles/ℓ moles/ℓ = gmw x 10^{-6}/ℓ = μ/ℓ) (Yalkowsky et al. 1983). The physical properties of the Aroclors fall within these ranges (Callahan et al., 1979; Mabey et al., 1981).

PCBs will volatilize from ambient waters with half-lives ranging between 2 months and >150 years based on an EXAMS model (Burns et al., 1981) using several published input parameters (Doskey and Andren, 1981; Callahan et al., 1979; Mabey et al., 1981). The most sensitive input parameter affecting volatilization rates was found to be the octanol/water partition coefficient (and those partition coefficients calculated from it, e.g., sediment, organic content and biota), showing that the partitioning of the PCBs from the sediments and biota onto the water was the limiting factor affecting volatilization.

Adsorption appears to be the dominant removal mechanism for highly chlorinated PCBs. Based on an EXAMS model (Burns et al., 1981) using the input parameters from the previously mentioned sources, sorption to sediment will effectively bind between 45 and >99% of the PCBs present in water depending on the PCB and the organic matter present in the sediment. The higher the organic matter in the sediment or the higher the chlorination, the more strongly sorbed was the PCB.

As for volatilization and sorption, biodegradation is also a significant removal mechanism for the less chlorinated species (Callahan et al., 1979; Hutzinger et al., 1972; Kaiser and Wong, 1974; Tabak et al., 1981). Based on published reports, PCBs containing ≤3 chlorines will be degraded in the environment, although estimation of half-lives is very difficult given the great variability in the reported literature. PCBs with four chlorines appear to be somewhat degradable, falling into middle ground. PCBs with five or more chlorines appear to be recalcitrant although a recent report (Brown et al., 1984) shows both aerobic and anaerobic degrading capabilities of microbial populations present in Hudson River sediments.

III. TOXICOKINETICS

Absorption

Gastrointestinal Absorption. One of the problems associated with understanding the toxicokinetics of PCB products is that they are mixtures of many different isomers and congeners, each with its own characteristic kinetics for absorption, distribution, metabolism, excretion and bioaccumulation. Fortunately, some of the studies on the toxicokinetics of PCBs have been done with relatively pure isomers, demonstrating that the degree and position of chlorination of the biphenyl moiety profoundly affect distribution and metabolism.

An early study of the oral absorption of 19 pure isomers, and the totally unchlorinated biphenyl was performed by Albro and Fishbein (1972). Male CD rats (number not reported) were gavaged with 5, 50 or 100 mg PCB isomer/kg bw (vehicle unspecified). Feces were collected for 4 days. The total amount of PCB excreted in the feces was determined and the percentage absorbed or metabolized was calculated from the amount administered minus the amount excreted. The results are presented in Table III-1. For all compounds tested at all dosage levels, absorption exceeded 90%. The authors recognized that this study did not investigate the possibility of intestinal metabolism, enterohepatic recirculation or the effects of dietary status on retention of PCBs, all of which would be required for full elucidation of the PCB absorption process.

Absorption was also determined by Allen et al. (1974a,b) who gave adult rhesus monkeys single oral doses of 1.5 or 3.0 g Aroclor 1248/kg bw (vehicle unspecified). Animals were killed and examined 4 days later. These authors also found that over 90% of the dose given had been absorbed as determined

TABLE III-1

Uptake of PCBs by Rats[a]

Compound	Percentage Retained[b]					
	5 mg/kg	± S.D.	50 mg/kg	± S.D.	100 mg/kg	± S.D.
Biphenyl	99.5	0.11	98.8	0.25	98.2	0.30
2-Cl	98.9	0.45	98.0	0.30	98.4	0.15
3-Cl	98.0	1.01	96.6	0.10	97.3	0.40
4-Cl	97.3	1.73	96.3	0.40	96.4	0.42
2,6-Cl_2			95.0	0.96	94.9	0.75
2,2'-Cl_2	95.0	2.42	97.4	0.20	98.2	2.10
2,4-Cl_2			95.7	0.40	97.1	0.35
2,3'-Cl_2			96.8	0.51	97.9	0.28
2,4'-Cl_2	97.2	3.38	95.5	1.20	96.6	0.78
3,4'-Cl_2	95.7	3.45	94.3	0.86	97.1	0.63
4,4'-Cl_2	95.4	3.05	95.6	1.12	97.4	1.10
2,5,2'-Cl_3			95.1	3.00	94.3	3.18
2,3,5'-Cl_3			95.6	1.05	95.0	1.12
3,4,3'-CL_3			93.5	2.85	90.3	3.58
2,4,4'-Cl_3			92.6	0.86	94.0	1.50
2,5,2',5'-Cl_4	96.0	1.75				
2,3,4,5-Cl_4	97.3	0.86				
3,4,3',4'-Cl_4	98.8	0.57				
2,4,5,2',5'-Cl_5	95.1	0.57				
2,4,5,2',4',5'-Cl_6	95.3	1.11				

[a] Source: Adapted from Albro and Fishbein, 1972

[b] Each value is the average of three determinations on each of two rats.

$$\text{Percentage Retained} = \frac{(\text{amount fed}) - (\text{amount excreted})}{\text{amount fed}} \times 100\%$$

by mass balance from the GI tract. Moderate hepatic enlargement, gastric mucosal hyperplasia and focal ulceration were prominent lesions. At the end of 4 days, tissue levels of PCBs were highest in the liver>brain>kidney from animals given either dose of Aroclor 1248. Animals sacrificed at 14 days postexposure showed liver levels greater than kidney. PCBs were not detected by gas chromatography in the brains. These authors confirmed the work of Albro and Fishbein (1972) that >90% of PCBs are absorbed from the GI tract.

In a subsequent experiment, Allen et al. (1975) administered one dose of 18 mg 2,5,2',5'-TCB/kg bw (vehicle unspecified) to seven adult male rhesus monkeys. Feces and urine were collected and analyzed for 14 days. The greatest amount of unmodified 2,5,2'5'-TCB was eliminated by the alimentary route on days 2-5. Over 12% of the original dose was recovered. Norback et al. (1978) administered one dose of ^{3}H-labeled 2,4,5,2'4'6'-HCB (vehicle unspecified) to two young monkeys with bile duct and duodenal canulae. The authors claimed that 59.3-87% of the total dose passed unabsorbed through the intestine within the first week. It was unclear why relatively little of this isomer was absorbed.

Dermal Absorption. Little pertinent information concerning dermal absorption of PCBs is available. Miller (1944) reported painting the skin of guinea pigs with 34.5 mg for 11 days with a "mixture" (undiluted) of isomers of diphenyl chlorinated in different positions and extent, with an approximate chlorine content of 42%, and an approximate empirical formula of $C_{12}H_3Cl_3$." All animals died by day 21 with lesions in liver, kidney, lungs, adrenals and heart that resembled those produced by parenteral injection. Doses of 86 mg of the previously described mixture every other day for 7 doses and 172 mg every other day for 8 doses to the skin of 11 rabbits resulted in death after 17-98 days. The number of doses varied from 9-15. Major lesions were fatty degeneration and centrolobular hepatocellular atrophy of the liver. The occurence of death and internal lesions typical of PCB intoxication is highly suggestive of considerable dermal absorption of PCBs. No attempt was made to quantitate the absorption of this PCB mixture.

Nishizumi (1976) reported on the dermal administration of ^{3}H-labeled Kanechlor-400 to nine male Wistar rats. A single dose of 0.001 mg [^{3}H]-Kanechlor-400 in olive oil was applied to a 1 cm diameter area of the clipped backs of rats. Rats were killed at days 1, 3 or 6 postexposure and ultramicroradiography was done. The author determined that dermal absorption was primarily through the hair follicle rather than through the stratum corneum. Appearance of "block curbicues" in dermal capillaries and hepatocytes lent credence to the thought that vascular distribution and hepatic accumulation occurred. No attempt was made to quantitate the amount of the dermally-applied dose that was absorbed.

Inhalation. Brief reports concerning inhalation exposure were found. Benthe et al. (1972) exposed male Wistar rats to one exposure of Pydranil A200 as an aerosol at a level of 30 g/m^3 (0.5-3 μ particles). Animals were killed and examined at 15 minutes, 2 hours, 24 hours and 2 days. At the end of 15 minutes, liver concentrations were >50% of maximum obtained after 2 hours. During the first 24 hours, rapid increase in brain and fat levels of PCBs were noted. At the end of 2 days, liver and brain levels had decreased from that observed at 24 hours, and adipose tissue levels of PCBs peaked. Therefore, absorption from inhalation exposure readily occurs.

Distribution

Distribution of PCBs following a single i.v. injection or gavaged dose was initially to liver and muscle, and eventually to adipose and skin. This phenomenon can best be described by discussing first those papers which deal with distribution of pure isomers. These data are summarized in Table III-2.

Studies in Rats. Matthews and Anderson (1975b) exposed groups of 13 male Sprague-Dawley rats to single doses of 0.6 mg of 4-[4C^{14}]-labeled PCB isomers. Levels of these isomers in blood, liver, skin, muscle and adipose at time intervals ranging from 15 minutes to 42 days were measured. Each of the PCBs was removed rapidly from the blood and distributed to liver and muscle. The large amount of PCB stored in muscle reflected its relatively large mass, >50% of body weight. Over a period of 7 days, the greatest concentrations were found in fat followed by skin. The fact that levels of 4-MCB were less in fat and liver than in muscle probably reflects that relatively rapid metabolism of this isomer results in its rapid excretion from the body. Giving the same dose of these isomers by oral gavage rather than i.v. resulted in markedly lower levels of all isomers (except 2,4,5,2',4',5'-HCB) stored in fat. The authors suggested that this may reflect increased initial metabolism of these compounds associated with oral absorption and initial hepatic distribution by portal circulation.

TABLE III-2

Tissue Distributions of Pure PCB Isomers Given to Animals

Species/Strain	Sex/Number	Source of PCBs	Vehicle	Route of Administration/Dosage	Tissues with Highest Concentration	Reference
Rat/Sprague-Dawley	M/13/group	[4-^{14}C]4-MCB	Emulphor	0.6 mg/kg bw i.v. once	1 hour: skin>fat>blood>muscle>liver 7 days: muscle>fat~skin>liver~blood	Matthews and Anderson, 1975b
		[4-^{14}C]4,4'-DCB			1 hour: fat>skin>liver>blood>muscle 7 days: fat>skin>liver>muscle>blood	
		[4-^{14}C]2,4,5,2',5'-penta-CB			1 hour: muscle>liver~fat>skin>blood 7 days: fat>skin>muscle>liver>blood	
		[4-^{14}C]2,4,5,2',4',5'-HCB			1 hour: muscle>liver>skin>fat~blood 7 days: fat>skin>>muscle>liver>blood	
Rat/Sprague-Dawley	M/3/group-time	[4-^{14}C]2,4,5,2',5'-penta-CB	Emulphor	0.6 or 6.0 mg/kg bw i.v. once	Initially: liver and muscle Redistribution: fat and skin	Matthews and Anderson, 1975a
Rat/Sprague-Dawley	M/2/group	[^{14}C]2,4,5,2',4',5'-HCB	Emulphor	0.6 or 3.6 mg/kg bw once by gavage or i.v. or gavage once weekly for 6 weeks	40 weeks: fat>excretions>skin>muscle>liver 6 weeks: fat>excretions>skin>muscle>liver	Muehlebach and Bickel, 1981
Rat/Sprague-Dawley	M/3/group	[^{3}H]2,5,2',5'-TCB	corn oil	50 mg by gavage	1 day: fat>stomach>blood>kidney>large intestine>skin>liver	Van Miller et al., 1975
Monkey/rhesus	M/F	2,5,2',5'-TCB	corn oil	0 or 18 mg once	2 weeks: fat>adrenals>>lungs~heart>liver~skin	Allen et al., 1975
Monkey/rhesus	M/2, F/2	[^{14}C]2,5,4'-tri-CB	fruit	550 μg/day for 84 days	Female: bile>fat>lymph nodes>ovaries>adrenal>liver Male: bile>fat>optic nerve>mammary gland>liver>adrenal After 28 days recovery, all tissues very low except bile, liver, skin, adrenal cortex, mammary gland of male.	Felt et al., 1977
Mice/NR	NR/NR	[^{14}C]2,4,5,2',5'-penta-CB	DMSO or intralipid	7 or 15 mg/kg bw once	Radioactivity rapidly leaves blood, taken up by liver, kidneys, brown fat. Redistribution: levels in fat increase; levels in other tissues decrease.	Berlin et al., 1975

Matthews and Anderson (1975a) administered 0.6 or 6.0 mg [4-^{14}C]-2,4,5,2',5'-penta CB/kg bw i.v. to male Sprague-Dawley rats. Tissue samples from three rats were taken at 2 and 8 hours and 1 and 4 days. Initially, liver and muscle accounted for most of the initial dose. Redistribution resulted in the majority of the initial dose located in adipose tissue and skin. In further studies on pharmacokinetics of PCBs, Anderson et al. (1977) and Lentz et al. (1977) substantiated their previous findings of initial distribution of these isomers to muscle and liver accompanied by a rapid decline in blood levels following an i.v. dose. Redistribution to fat and skin was confirmed.

Male Sprague-Dawley rats (2/group) were treated with a single dose of 0.6 or 3.6 mg [^{14}C]-2,4,5,2',4',5'-HCB/kg bw by gavage or i.v. injection (Muehleback and Bickel, 1981). Another group was treated by gavage once weekly for 6 weeks. The first groups were examined after 1 hour, 24 hours, 6, 20 or 40 weeks. Initially muscle, liver, fat and skin contained the highest levels of the PCB. Redistribution rapidly resulted in adipose tissue containing the most PCB, followed by skin, muscle and liver. Excretions accounted for only 16% of the total dose at the end of 40 weeks. These tissue distributions resembled those reported by Matthews and Anderson (1975a,b). The authors concluded that 2,4,5,2',4',5'-HCB is resistant to metabolism and accumulates in body fat.

Treatment of groups of three male Sprague-Dawley rats with one dose of 50 mg [^{3}H]-2,5,2',5'-TCB by gavage demonstrated the metabolizeable nature of this isomer (Van Miller et al., 1975). After 1 day, tissue levels were highest in fat>stomach>blood>large intestine>skin>liver. At the end of 14 days, fecal (72.5%) and urinary (12.9%) radioactivity indicated the rapidity with which this isomer was being metabolized and removed. Since skin seems to be a depot for relatively non- or more-slowly metabolized isomers, it was not surprising to see skin levels of 2,5,2',5'-TCB comparatively low. At the end of 7 days, all tissue levels of the PCB were very low.

Studies in Mice. Berlin et al. (1975) reported on the i.v. administrations of a single dose of 7 or 15 mg [^{14}C]-2,4,5,2',5'-penta-CB to mice. Scintillation counting of tissues and whole body autoradiography revealed that radioactivity rapidly left the blood and was taken up by liver, kidneys and brown fat. Within 16 days, redistribution had caused PCB levels in fat to increase markedly while other tissue levels became very low.

In a more sophisticated protocol, Gage and Holan (1976) administered 14 isomers of PCBs to mice at levels of 13-165 μg/mouse by single dose gavage. Concentrations in abdominal fat were determined at 7 and 21 days (Table III-3). Especially high levels in fat were noted of 2,4,5,2',4',5'-; 4,2'4'6'- and 2,4,2'4'-PCBs after both 7 and 21 days.

Studies in Primates. Similarly, Allen et al. (1975) treated four male rhesus monkeys with 18 mg 2,5,2'5'-TCB by gavage. Relative tissue levels at the end of 14 days were fat>>adrenals>>lungs~heart>liver~skin. At the end of that time, 12% of the original dose had been recovered from the feces, indicating very high absorption.

TABLE III-3

Retention of PCBs in Mouse Fat at 7 and 21 Days After Dosing[a,b]

Chlorine Biphenyl Positions	Dose (μg/mouse)	Concentration in Fat (ng/g/μg dose)					
		7 Days			21 Days		
		Group Size	Mean	SD	Group Size	Mean	SD
4,4'	20	6	<10		6	<10	
2,4,2'4'	25	6	100	8	6	41	11
2,4,2',5'	25	6	28	13	6	26	15
2,5,2',5'	25	6	54	13	6	20	6
2,5,3',4'	25	6	15	10	6	<10	
3,4,3',4'	25	6	27	8	6	<10	
3,5,3',5'	25	6	77	12	6	<10	
3,2',4',6'	26	6	<10		6	>10	
4,2',4',6'	25	6	108	11	6	70	16
2,5,2',4',5'	165	3	32[c]	11	3	25[c]	8
2,3,2',4',6'	28	6	<15		6	<15	
2,3,4,2',4',5'	31	6	178	62	6	160	46
2,3,4,2',4',6'	13	3	<10		4	<10	
	13	3	17[d]	8	4	14[d]	1
2,4,5,2',4',5'	31	4	117	15	4	132	24
	31	4	108[d]	16	4	131[d]	21

[a] Source: Adapted from Gage and Holm, 1976

[b] All analytical determinations were performed by gas-liquid chromatography on days 7 and 21 except where noted.

[c] Radioactive assay 8 and 16 days after dosing

[d] Radioactive assay

Tri-CB in monkeys is also rapidly metabolized and removed. Felt et al. (1977) gave two male and two female rhesus monkeys 550 mg [^{14}C]-2,5,4'-tri-CB in a piece of fruit daily for 84 days. Necropsy of one male and one female revealed greatest concentrations in bile and fat. The female also had high levels in lymph nodes, ovaries, adrenals and liver. The male had high levels in the optic nerve, mammary glands, liver and skin. After 28 days of recovery, the other two were necropsied. All tissues at that time were low except for the bile, liver, skin, adrenal cortex and the mammary gland of the male. Relatively rapid metabolism of this isomer resulted in fairly low levels in the skin.

Studies on PCB Mixtures to Rodents. Administration of PCB mixtures to rodents have also been studied. These studies are summarized in Table III-4. Hashimoto et al. (1976) reported on the administration of [^{14}C]-Kanechlor-400 or [^{14}C]-Kanechlor-600 to groups of 12 JCL-S.D. rats by gavage for 5-50 weeks. Total dose of PCBS received ranged from 6.35-7.85 mg. Redistribution resulted in the highest levels in fat>skin>adrenals> aorta>sciatic nerve>blood. Concentrations in other tissues were low.

Kurachi and Mio (1983b) exposed groups of 4-10 ICR mice to 100 mg Kanechlor-400/kg diet for 5-20 days. Analysis of tissue distributions of PCBs at the end of the feeding period indicated high levels of PCBs in the gonads. Other tissues with significantly high levels of PCBs were skin, adipose tissue, adrenals and kidneys. Mice were kept on the PCB diet for 20 days in a rotating cage to prevent resting and cause fatigue. Mobilization

TABLE III-4

Tissue Distributions of PCB Mixtures Given to Animals

Species/ Strain	Sex/ Number	Source of PCBs	Vehicle	Dosage/Route of Administration	Tissues with Highest Concentration	Reference
Rat/JCL-S.D.	M/12/ group	$[^{14}C]$ Kanechlor 400 or $[^{14}C]$ Kanechlor-600	olive oil	totals of 0, 7, 14, 7, 17, 7.64, 7.87 or 6.35 mg/kg bw over 0, 5, 10, 30, 50 or 5 weeks, respectively	Fat>skin>adrenal> aorta>sciatic nerve> blood	Hashimoto et al., 1976
Mice/ICR	NR/4-10/ group	Kanechlor-300, -400 or -500	diet	100 mg/kg diet for 5-20 days	Gonad>skin>fat> liver>adrenals> kidney	Kurachi and Mio, 1983b

of fat deposits occurred with a corresponding release of stored PCBs. Mice so treated had liver levels of PCBs 10 times greater than mice subjected to the same diet but allowed to rest.

Distribution to the Fetus and Neonate (Rodents). PCBs can cross the placenta and accumulate in fetuses. PCBs are also excreted by lactating dams through the fat portion of milk. PCBs can cause a high incidence of fetal toxicity and accumulate in high levels in the tissues of nursing young, it is therefore appropriate to discuss these phenomena separately from general tissue distribution. These studies are summarized in Table III-5.

Torok and Weber (1981) administered tracer amounts of [^{14}C]-2,4'5'-tri-CB or a mixture of [^{14}C]-PCBs by gavage to pregnant NMRI mice on days 13, 15 and 17 of pregnancy. Dams were killed and analyzed by scintillation counting of tissues and autoradiography on days 14, 16 or 18. Highest levels of labeled tri-CB in dams treated with tri-CB were in fat. Levels in mammary gland were also very high followed by levels in the liver, kidney and ovary or blood. Tissue levels of the PCB mixture ([^{14}C]-PCB) were highest in fat followed by liver, mammary gland and kidney. All tissue levels of the PCB mixture were lower than levels of tri-CB. Tissue levels of tri-CB and the PCB mixture in the conceptus were comparable on all 3 days of examination. Both fetal and placental levels of tri-CB and the mixture increased from day 14 to day 16. The most dramatic increase occurred on day 18. Fetuses on day 18 showed levels 2-5 times higher than they did on day 14. Autoradiography of fetuses revealed fairly uniform organ distribution

TABLE III-5 (cont.)

Species/Strain	Sex/Number	Source of PCBs	Vehicle	Dosage/ Route of Administration	Distribution to Dams Milk or Offspring	Reference
Rat/Sprague-Dawley	F/NR	Aroclor 1254	diet	0, 25 or 50 mg/kg diet starting day 8 of gestation - day 14 postpartum	Dam: fat>mammary>kidney>liver>lung Milk: postpartum day 0/293 mg/mℓ; postpartum day 14 (32 μg/mℓ).	McCormack et al., 1979
Monkey/rhesus	F/16	Aroclor 1248	diet	0, 2.5 or 5.0 mg/kg diet continuously 1.5 years, start 6 months prebreeding	Stillborn male-5.0 mg/kg: lung>pancreas>adrenal>thymus>spleen>muscle>kidney>liver. Male died at 44 days-2.5 mg/kg: bone marrow-lung>thymus>adrenal>pancreas>kidney>spleen.	Allen and Barsotti, 1976
Monkey/rhesus	F/4	Clophen A-30	1% methyl cellulose in water	0 or 16 mg/kg bw/day by gavage 22-29 days	Blood of offspring higher in PCBs than blood of dams. PCB levels in milk 10-20 times that of serum of dam. Higher PCB levels in tissues of infant than dam.	Iatropoulos et al., 1978; Bailey et al., 1980

TABLE III-5

Distribution of PCBs to Fetuses and Nursing Young

Species/Strain	Sex/Number	Source of PCBs	Vehicle	Dosage/ Route of Administration	Distribution to Dams Milk or Offspring	Reference
Mice/NMRC	F/NR	$[^{14}C]$2,4',5'-tri-CB $[^{14}C]$ Mixture of PCBs	sesame oil	trace amounts/gavage on days 13, 15, 17 of pregnancy	In dams: fat>mammary gland>liver>kidney> ovary-blood. Fetal: day 18 levels>14, mostly in GI tract and liver at day 18.	Torak and Weber, 1981
Mice/ddN	F/10-15/ group	2,4,4'-tri-CB; 2,4,5,3'-tetra-CB; 2,4,5,2'5'-penta-CB; 2,4,5,2',4',5'-hexa-CB; 2,3,4,2',4',5'-hexa-CB; 2,3,4,5,6,2',5'-penta-CB or 2,3,4,5,2',3',4',5'-octa-CB	diet	0.32 mg/kg diet 0.42 mg/kg diet 0.44 mg/kg diet 0.44 mg/kg/diet 0.16 mg/kg diet 0.23 mg/kg diet	Fetal levels: greater when PCB fed during rather than before pregnancy. In all cases tested, exposure of dams pregestational elevated fetal tissue levels.	Masuda et al., 1979
Mice/ddN	F/7-12/ group	Kanechlor-500	diet	0.01 (control), 0.94, 8.4 or 86 mg/kg diet from days 0-18 of pregnancy or day 0 of pregnancy to 5 weeks postpartum	Fetuses: whole body levels dose-related; at 86 mg/kg diet level fetal adipose tissue PCB >maternal liver PCB concentrations. PCB levels of offspring during lactation ≥100x previous fetal levels.	Masuda et al., 1978
Mice/NMRI	F/21 F/25	2,4'5-tri-CB or 2,2',4,4',5,5'-HCB	peanut oil	0 or 0.05 mg/days for days 5-19 of gestation 0 or 0.05 mg/day for days 1-12 of lactation	Dams: liver HCB>>tri-CB Fetuses: adipose HCB>>tri-CB Dams: liver HCB>>tri-CB Offspring: liver HCB>>tri-CB; stomach contents HCB>>tri-CB	Orberg, 1977
Rat/Sherman	F/6.13/ group	Aroclor 1254	peanut oil	0, 10 or 50 mg/kg bw/day, days 7-15 of gestation	Fetuses: <0.12, 0.63, 1.38 μg/g tissue Milk: <0.75, 20.63, 66.2 μg/g milk 21-day-old weanling males: 0.19, 3.35, 18.24 μg/g liver	Curley et al., 1973
Rat/Wistar	F/5/group	Aroclor 1254	drinking water	6.4 mg/kg bw daily for 9 weeks	Fetal levels: low: ~1 μg/g whole body 10- and 20-day-old nursing pups: 10-80+ μg/g whole body	Baker et al., 1977
Rat/Wistar-SLC	F/12/group	Kanechlor-400	diet	0, 10, 50 or 250 mg/kg diet during gestation	PCB concentrations in fetus >blood of dams. Marked increase at day 14 of gestation. PCB concentrations in 22-day-old offspring >tissue levels of dams. Tissue levels of 28-day-old pups whose dams received 50 mg/kg: liver>kidney> lung>spleen>heart>brain.	Mizunoya et al., 1974

of PCBs on day 14. By day 18, radioactivity was concentrated in the GI tract and liver. Fetal content of labeled PCBs exceeded maternal blood levels at this time.

Masuda et al. (1979) examined the transfer of seven PCB isomers to the fetuses and offspring of pregnant ddN mice exposed for the first 18 days of pregnancy or 18 days immediately before pregnancy. There were 10-15 mice in each group and levels of PCB ranged from 0.16-0.44 mg/kg diet. This fairly complicated protocol is summarized in Table III-5. Masuda et al. (1979) found little transfer of PCBs to the fetus, comparable to the levels observed by Torok and Weber (1983). Maximum PCB levels occurred in offspring 1-2 weeks old, undoubtedly as a result of nursing. Levels of PCBs in milk from lactating mice were seen to diminish reapidly after ~2 weeks. Tissue levels of offspring declined markedly after 3 weeks when the pups were weaned and thus no longer exposed to PCBs in their mothers milk. The researchers found that those isomers which transferred more readily to the fetus generally transferred to the offspring by lactation. The two hexa-CB isomers transferred most readily. The tri- and octa-isomers transferred in next greater quantity and tetra- and penta-isomers transferred the least. The hepta-isomer transferred so poorly that it was often totally undetectable, especially in fetuses and nursing or weaned offspring. It was noted that the position of chlorination may have been more important than degree of chlorination in explaining tissue levels in offspring. PCBs having unsubstituted hydrogen atoms at positions 3 and 4, such as 2,5,3',4'-tetra-CB, 2,4,5,2',5'-penta-CB and 2,3,4,5,6,2',5'-hepta-CB accumulated less in dams and offspring.

In an earlier study, Masuda et al. (1978) fed diets containing 0.01 (control) or 86 mg Kanechlor-500/kg diet to pregnant ddN mice from day 0-18 of gestation. Fetal whole body levels of PCB were low but dose-related. Concentration of fetal adipose PCBs exceeded maternal hepatic concentrations when dams were fed the high (86 mg/kg diet) level. After 5 weeks of lactation, concentration of PCBs in offspring was ≥100 times previous fetal concentrations, emphasizing the ability of many PCBs to transfer to offspring by lactation.

Orberg (1977) also determined that 2,2',4,4',5,5'-HCB crossed the placental barrier more efficiently and was found in milk of nursing mice in greater concentration than 2,4'5-tri-CB. NMRI mice were given 0 or 0.05 mg of either isomer once daily on days 5-19 of gestation or days 1-12 of lactation. At the conclusion of each feeding period the dams and progeny were killed and examined for levels of PCBs. This author confirmed the findings of Masuda et al. (1979) that the 2,2',4,4',5,5'-HCB isomer crosses the placental barrier and the mammary gland barrier more efficiently than does the 2,4',5-tri-CB isomer. He also directly measured PCB levels in milk (stomach contents of sucklings after a meal) and found the hexa-CB isomer present in much higher concentration than the tri-isomer.

Various PCB mixtures (commercial products) have been given to rats to study transplacental passage and transfer by lactation. Curley et al. (1973) gave 10 or 50 mg Aroclor 1254/kg bw orally to Sherman rats on days 7-15 of gestation. Fetuses were examined on day 20 of gestation. Pups were found to have low but dose-related concentrations of PCB-derived components. Milk was taken from stomachs of nursing rat pups 11 days after the last dose was given to the dams. Substantial levels of PCB-derived components were found. Concentrations of PCB-derived components in livers of 21-day-old weanling males were 5- to 10-fold greater than whole-body levels in 20-day-old fetuses. This indicates the passage of PCBs into milk, and the consequently higher exposure of nursing pups than gestating fetuses to PCBs administered orally to the dam. Because the increase in fetal PCB-derived components from the 50 mg/kg/day dosed dams was only 2-fold and not 5 times the concentration of PCB-derived components from the 10 mg/kg/day dosed dams, the authors concluded that the placenta may constitute a partial barrier to the transfer of PCB-derived components.

By providing 6.4 mg Aroclor 1254/kg bw in the drinking water for 9 weeks, Baker et al. (1977) were able to induce fetal resorption in pregnant Wistar rats. In this experiment, which was designed primarily to study long-term PCB-induced toxicity, these workers found fetal concentration of PCBs approximating 1 μg/g of whole body. These results agree with the findings of Curley et al. (1973), who determined whole body concentrations of pups after 10-20 days of nursing to be ~10-80 μg/g tissue and also concluded that the placenta presents at least a partial barrier to passage of PCBs.

Mizunoya et al. (1974) fed diets containing 10, 50 or 250 mg Kanechlor-400/kg of diet presumably during gestation and lactation to groups of 12 Wistar-SLC rats. They noted that PCB concentrations in fetal tissues were lower than concentrations in maternal blood. They also confirmed the results of the previously discussed studies that tissue levels in 28-day-old pups exceeded PCB levels in maternal tissues. Tissue levels in 28-day-old weaned pups were greatest in liver>kidney>lung>spleen>heart>brain in offspring of dams exposed to 50 mg PCB/kg diet. Concentrations in skin were not determined.

PCBs in the diet of Sprague-Dawley dams produced tissue concentrations greatest in the adipose>mammary>kidney>liver>lung (McCormack et al., 1979). Aroclor 1254 was administered at 0.25 or 50 mg/kg diet starting on day 8 of gestation and continuing through postpartum day 14. The most remarkable finding was the concentration of PCBs in milk, 293 μg/mℓ by postpartum day 14.

Distribution to the Fetus and Neonate (Primates). The transplacental and mammary transfer of PCBs in monkeys has been of special interest because infants nursed by PCB-exposed dams develop signs of PCB-induced intoxication rather quickly. Allen and Barsotti (1976) fed diets containing 2.5 or 5.0 mg Aroclor 1248/kg diet starting 6 months before breeding 16 female rhesus monkeys. PCB exposure continued for about 18 months. One male infant was stillborn, reportedly from dystocia. PCB concentration in its tissues ranged from 99.5 μg/g in lung>pancreas>adrenal>thymus>spleen>muscle> kidney>2.5 μg/g in liver. Surviving infants all developed typical facial skin lesions by 2 months of age. Tissue levels of PCBs in a male infant that had died at 44 days of age ranged from 50.8 μg/g in bone marrow ~lung>thymus>adrenal>pancrease>kidney to 0.62 μg/g in liver. The authors concluded that PCBs received from nursing resulted in clinical signs of intoxication in monkey offspring.

Iatropoulos et al. (1978) and Bailey et al. (1980) treated three nursing rhesus monkeys with 16 mg Clophen A-30/kg bw/day by gavage starting on days 46-127 postpartum and continuing for 22-29 days. The youngest infant and her dam became moribund on exposure day 21 and were killed. Necropsy of the infant revealed mild hepatocellular pathology and dilatation of renal tubules containing casts. The most striking lesions consisted of moderate gliosis and demyelination of the central nervous system. Levels of PCBs in milk appeared to range from 10-20 times the levels in serum. In general, serum levels of PCBs in infants were about 2 times the levels in their dams. Tissue levels of PCBs in infants varied widely. Body fat contained the highest levels (1687 μg/g)>bone marrow>lymph nodes>adrenals>thymus> kidney>spinal cord>liver (80 μg/g). In all tissues except the thymus, infant tissue levels were 1.94-5.47 times the corresponding levels in the moribund dam sacrificed after 22 days of exposure. It was concluded that lactation greatly concentrates PCBs, and consequently, exposes nursing young to greater levels than the dam herself. The nursing infant is, therefore, an individual at special risk to PCB intoxication.

Strong evidence exists that lactation is a route of exposure for human infants as well as offspring of animals. Miller (1977) reported an analysis of 80/1600 samples of human breast milk collected in 1974-1975 by the EPA to evaluate pesticide content. Of them, 16 had levels of PCBs ranging from 2.5-10.6 μg/mℓ when measured from 16-540 days after delivery. Wolff (1983) reports the half-life of PCBs in breast milk as 5-8 months, and that the concentration of PCBs in breast milk is 4-10 times that in maternal blood.

Rogan et al. (1980) examined 1083 samples of human breast milk collected in 1975. They found that 30% had levels $\geq$50 µg/mℓ, with a range of 50-409 ng/mℓ. Wickizer et al. (1981) examined breast milk from 1057 samples collected in Michigan from 1977-1978. The mean level was 1.496 µg/mℓ and 6.14% with >3 µg/mℓ. Mes and Davies (1979) examined breast milk of Canadians. They reported 98% of samples contained at least 1 ng/g whole milk. Kuwabara et al. (1979) measured PCBs in blood in children and mothers who were nonoccupationally exposed to PCBs in Japan. Data from 16 mothers and 17 breast-fed children were obtained. Blood levels of PCBs of mothers' blood averaged 2.8 ng/mℓ; levels in children averaged 3.8 ng/mℓ. In general, longer nursing periods increased blood levels of PCBs in children indicating that lactation is a major route of PCB excretion. Hofvander et al. (1981) reported on levels of PCBs in breast milk collected from mothers at 3 months (18 samples) and 6 months (23 samples) postpartum. Mean PCB levels were 65 (15-150) and 63 (19-120) ng/g milk after 3 and 6 months of nursing, respectively. No differences were seen in PCB concentrations between the 3- and 6-month samples.

Jacobson et al. (1984) report the transfer of PCBs across human placenta and into maternal milk. Maternal serum levels were significantly higher than cord serum levels ($p<0.001$). When estimated on a fat basis, however, PCB levels were not significantly different between maternal and cord serum. Mean PCB levels were also higher in maternal milk when compared to maternal serum: 19.3 ng/mℓ vs. 4.7 ng/mℓ, respectively.

Bioaccumulation and Retention

The discussion of distribution pointed out that an initial dose of PCBs is readily absorbed and transferred rapidly from the bloodstream to various body tissues. Muscle tissue because of its mass, and hepatic tissue because of its central location for circulation and for metabolism seemed to be the initial loci of distribution. Redistribution to body depots occurred rapidly as PCBs left muscle tissue and other organs. Adipose tissue appeared to be the locus for long-term bioaccumulation and retention. Other fat and lipid containing tissues, such as brain, bone marrow and adrenal, are also likely to be high in PCBs.

In the studies by Matthews and Anderson (1975a,b), it was shown that 7 days after a single dose of four PCB isomers body retention was primarily in fat and skin (see Distribution Section). They found that 4-MCB did not bioaccumulate in body fat, probably because of its rate of metabolism and removal. The more heavily chlorinated isomers and those that tend to metabolize more slowly are those that would persist in body tissues such as fat and skin.

Muehlebach and Brickel (1981) administered single i.v. doses of 0.6 or 3.6 mg 2,4,5,2',4',5'-HCB to a total of seven male Sprague-Dawley rats. Blood levels dropped rapidly as distribution occurred to muscle>liver>skin~fat in the first 24 hours. By 6 weeks redistribution was complete. PCB concentrations were highest in adipose and then skin, muscle and liver; concentrations were lowest in blood. By 40 weeks excretion had only accounted for 16% of the total dose of hexa-CB given for both the 0.6 and 3.6 mg/kg exposed rats.

Allen et al. (1975) found that rhesus monkeys tended to accumulate a single dose of 2,5,2',5'-TCB in body fat 2 weeks after exposure. For this isomer, storage in skin was not a major body depot. Large term administration of 2,5,4'-tri-CB to rhesus monkeys (Felt et al., 1977) resulted in the highest concentrations in bile, which was evidence of the relatively rapid rate of metabolism of this isomer. Adipose tissue was the body tissue containing the highest concentrations of the isomer.

Gage and Holm (1976) administered 14 separate isomers to mice and measured retention at 7 and 21 days. Retention in body fat of 2,4,5,2',4',5'-HCB was greatest. The 4,4'-DCB, 3,2'4'6'-tetra-CB and 2,3,4,2',4',6'-HCB isomers were retained the least, indicating that position of chlorination was as important as degree of chlorination in determining body retention.

Studies of bioaccumulation in humans has been conducted primarily on necropsy specimens. Kraul and Karlog (1976) reported an analysis of PCBs in abdominal fat, brain and liver from necropsy material collected in 1972 and 1973 in Copenhagen, Denmark. On a fresh weight basis, they reported ratios of 1:3.5:81 for PCBs in brain:liver:fat from 82 sets of organs. Adipose tissue was clearly the site of greatest bioaccumulation of the organs studied.

Yobs (1972) examined PCB levels in adipose tissue of necropsy specimens and surgery patients from 18 states and found 198/637 (31.1%) to be positive. All states represented had positive samples. Mes et al. (1977) reported on PCB analysis of adipose tissue of Canadians collected in 1972. They found Aroclor 1260 levels ranging from 0.106-6.603 μg/g fresh tissue.

Residues in residents of Ontario were highest (1.07 μg/g tissue), and residues in residents of central Canada were lowest (0.499 μg/g tissue).

Wasserman et al. (1978) reported on the residues of PCBs in neoplastic and adjacent, apparently normal tissue, from 17 human patients with gastric carcinoma, and 15 healthy patients, apparently free of gastric carcinoma. They found that the concentrations of PCBs in lipids extracted from tumor tissue (15.33 μg/g) were much higher than the concentration in apparently normal adjacent tissue (6.29 μg/g). Stomach tissues of cancer patients had higher concentrations of PCBs than stomach tissues of controls.

Metabolism

A discussion of the metabolism of PCB products is complicated by the fact that PCBs in the environment to which individuals may be exposed are likely to consist of a large number of individual isomers. Each PCB isomer has toxicodynamic characteristics which may vary from species to species. Several good reviews on the subject exist (Yoshimura and Yoshihara, 1976; Sundstrom et al., 1976a,b,; Safe, 1980). The purpose of this discussion will be to review the pertinent facts of metabolism for each individual isomer or group of isomers studied in sufficient detail to define major metabolic pathways. Modification of toxicity of the parent compound, when demonstrated, will be noted.

Generally, it seems that degree of chlorination greatly influences rate of metabolism; the more heavily chlorinated the biphenyl moiety, the slower the metabolism. Metabolism of isomers containing four or more chlorine atoms may be influenced more by position of chlorination than degree of chlorination (Safe, 1980). Since the mono-, di- and trichlorinated isomers behave similar metabolically, they will be discussed first.

Metabolism of Mono-, Di- and Tri-chlorinated Isomers. Hass et al. (1977) treated male Sprague-Dawley rats with 30 mg [^{14}C]4-CB or [^{14}C]-4,4';-di-CB/kg bw by gavage in corn oil for 7 or 6 days, respectively. Administration of 4-CB resulted in the formation of four metabolites: 4-hydroxy-4'-CB accounted for 65% of the radioactivity excreted in urine and feces; 3,4-dihydroxy-4'-CB accounted for 22% of excreted radioactivity; and either 3-methoxy-4-hydroxy- or 4-methoxy-3-hydroxy-4'-CB accounted for 2.5% of total excreted radioactivity. Less than 10% of the recovered radioactivity cochromatographed with the parent compound, which indicated that absorption and metabolism of 4-CB was over 90% complete. Injection of a 0.6 mg/kg bw dose of 4-CB into the femoral vein of bile-cannulated rats and recovery of hepatic metabolites revealed a half-life of only 3.6 hours for the parent compound. This work was confirmed by Hutzinger et al. (1972) who determined monohydroxylated-CB to be the major metabolite of 4-CB.

Safe et al. (1975a) administered one dose of 4-chlorobiphenyl by i.p. injection to a rabbit. Excreta were collected for 10 days and analyzed. Major urinary metabolites identified were 4-hydroxy-4'-CB and 3,4-dihydroxy-4'-CB. Further investigation of the formation of 4-hydroxy-4'-CB suggested the formation of an intermediate, arene oxide. Block and Cornish (1959) administered 1 g of 4-chlorobiphenyl by gavage to six male white rabbits. Analysis of urine collected 4 days before and after dosing with 4-CB revealed the formation of glucuronide conjugates as the major metabolite excreted through the urine. Fecal analyses were not performed. These authors (Safe et al., 1975b) administered 5 mg 4-CB/kg bw i.v. to two goats or 50 mg/kg bw in a gelatin capsule orally to a cow. Urine and feces were collected for 5 and 8 days, respectively. Analysis revealed urinary excretion of 4'chloro-4-biphenylol and 4'chloro-3,4-biphenyldiol. These metabolites were not found in feces.

In the study on rats described previously, Hass et al. (1977) determined the major metabolite of 4,4'-di-CB found in the bile to be 4-hydroxy-3,4'di-CB. This metabolite, which required a chlorine shift, constituted 91% of the di-CB excreted. A minor metabolite was 3-hydroxy-4,4'di-CB, which required no chlorine shift in its formation. Unmetabolized 4,4'-di-CB constituted <10% of the di-CB recovered from bile. Various monohydroxylated species were also reported by Hutzinger et al. (1972). Anderson et al. (1977) indicated that 3-hydroxy-4,4'-di-CB was the major metabolite of 4,4'-di-CB. In their study on urinary metabolites of PCBs in goats and a cow, Safe et al. (1975a) found only 3-hydroxy-4,4'di-CB. Safe et al. (1976b) administered 0.3 g 4,4'-di-CB in vegetable oil to a male rabbit and collected urine and feces for 7 days. Urinary metabolites determined were 3-hydroxy-4,4'-di-CB, 4-hydroxy-3,4'di-CB and 4-hydroxy-4'-CB. Formation of this later metabolite required dechlorination and subsequent hydroxylation.

Goto et al. (1974a) administered 20 mg [^{14}C]-2,3-di-CB orally to a rat and collected urine and feces for the next 10 days in 5-day collections. Unchanged di-CB was collected only during the first 5-day collection period.

It was isolated from the feces and constituted only 5.7% of the total dose of radioactivity given. Only one metabolite, 4-hydroxy-2,3-di-CB, occurred. In the urine it occurred exclusively as a glucuronide conjugate.

In a concurrent study, 23 mg of [^{14}C]-2,4,6-tri-CB was given orally to a rat, and metabolites were isolated from feces in urine over the next two periods of 5 days each. Slightly more of the tri-CB, 9.2%, was found unchanged in the feces. Equal amounts of radioactivity occurred in feces and urine at the end of the 10-day collection period. In urine, conjugates of two metabolites were found; one was 3'-hydroxy-4'methyl-tri-CB (or 3'-methyl-4'hydroxy-tri-CB) and the other was 3',4'-dihydroxy-tri-CB. Incubation of 2,5,2'-tri-CB with phenobarbital-activated rat hepatic microsomes resulted in the formation of monohydroxylated tri-CB metabolites (Ghiasuddin et al., 1976). As many as six separate metabolites resulted when tri-CB was incubated with microsomes. Uninduced rat hepatic microsomes did not result in any detectable metabolism of this tri-CB isomer. Using phenobarbital-induced microsomes resulted in metabolism of 67% of the tri-CB present. Fruit containing 550 mg of [^{14}C]-2,5,4'-tri-CB was fed to two male and two female monkeys for 84 days. Urine and feces were collected and extracted for metabolites. Because of the difficulty of finding sufficient pure metabolite in feces, isolation and quantitation were not possible. Urinary analysis revealed the presence of a glucuronide of 3-hydroxy-4,2'5'-tri-CB. Another minor metabolite, 4-hydroxy-2,5,4'-tri-CB was also present.

The rate of in vivo metabolism of the tetra-CB isomers depends to a large extent on the positions of chlorine atoms. Studies of the metabolism of two tetra-CBs have been found. Goto et al. (1974b) administered

[^{14}C]-2,3,5,6-tetra-CB to a rat and collected feces and urine for two consecutive 5-day periods. They were able to isolate two main fractions and determined the major metabolites to be 4-hydroxy-,3-hydroxy- or 3,4-dihydroxy-2',3',5',6'-tetra-CB. A fourth metabolite was determined to be 3-hydroxy-4-methoxy-2',3',5',6'-tetra-CB. In the urine, only conjugates of these metabolites occurred and accounted for 12% of the radioactivity recovered as metabolites. Fecal excretion accounted for 88% of the radioactivity recovered as metabolites. About 40% of the dihydroxy derivatives occurred as conjugates and 13% of the 3-hydroxy, 4-hydroxy- or 3-methoxy-, 4-hydroxy- derivatives (collectively) were conjugated.

Van Miller et al. (1975) administered 50 mg [^{3}H]-2,5,2',5'-TCB in corn oil to four groups of male Sprague-Dawley rats (3 rats/group). Feces and urine were collected at 24-hour intervals until the groups were killed at 1, 3, 7 or 14 days for tissue analysis. Extraction of metabolites from urine and feces followed by thin layer chromatographic separation revealed six separate metabolites of 2,5,7',5'-TCB. Definitive identification of each one was not possible. Over 90% of tritium recovered from feces occurred in a form other than the parent compound, indicating a high degree of absorption and metabolism. At least 60% of the metabolites were identified as monohydroxy derivatives. Dihydroxy derivatives were also postulated. Hydroxylation through epoxide intermediates was suggested. It was stated that the toxicity of TCB to rats can be modified by treatment with SKF-525A or phenobarbitol. Pretreatment with SKF-525A, which retards metabolism of xenobiotics, enhanced toxicity; and phenobarbitol pretreatment, which enhances metabolism of xenobiotics, reduced toxicity of TCB (unpublished data). Metabolism and consequent excretion, therefore, was considered to be a major factor in detoxification of PCBs.

Norback et al. (1976) subsequently administered 50 mg $[^3H]$-2,5,2',5'-TCB/kg bw to four male and four female Sprague-Dawley rats following bile duct cannulation. Bile was collected in 1 mℓ aliquots for 24 hours. Within the 24-hour collection period, 16% of the total radioactivity was recovered from the bile. The major metabolite recovered was 3-hydroxy-2,5,2',5'-TCB. A metabolite which had gas chromatographic retention time and mass spectra characteristics identical to *trans*-3,4-dihydro-3,4-dihydroxy-2,5,2',5'-tetra-CB was also found. The authors noted that the formation of *trans*-dihydrodiols was excellent evidence for the formation of arene oxides as intermediates. Enzymatic hydration of arene oxides yields *trans*-dihydrodiols.

Earlier, Gardner et al. (1973) had administered 10 mg of 2,5,2',5'-TCB to three male New Zealand White rabbits. Daily aliquots of urine were collected for 1 week. Urine was extracted, purified by thin layer chromatography and subjected to spectroscopic and chromatographic analyses. Comparison of the characteristics of the extracted metabolites with pure synthetic compounds allowed identification of the metabolites. The major urinary metabolites found were 3- or 4-hydroxy-2,5,2',5'-tetra-CB. A third metabolite was *trans*-3,4-dihydro-3,4-dihydroxy-2,5,2'5'-tetra-CB. The authors also noted the formation of *trans*-dihydrodiol as evidence for intermediate formation of the arene oxide 2,5,2',5'-tetra-CB-3,4-oxide. This intermediate could also lend to formation of 3- and 4-hydroxy metabolites.

Metabolism of 2,5,2',5'-TCB in the nonhuman primates probably proceeds by arene oxide intermediates but results in metabolites that differ at least in proportion from those found in the rat and rabbit. Hsu et al. (1975a,b) administered 500 mg $[^{3}H]$-2,5,2',5'-tetra-CB/kg bw in corn oil to four infant male (600-800 g) rhesus monkeys. Feces and urine were collected in 24-hour aliquots for 72 hours. Monohydroxy derivatives of TCB were found in the urine, but they constituted a much smaller part of the total radioactivity recovered as metabolites than was the case in the rat or rabbit. The major urinary metabolites were dihydroxy-tetra-CB and *trans*-3,4-dihydro-3,4-dihydroxy-tetra-CB. A second minor metabolite was identified as 3,4-dihydroxy-tetra-CB. The authors suggested that the preponderance of the *trans*-dihydrodiols and 3,4-dihydroxy derivatives strongly indicated the role of intermediate arene oxide formation in metabolism of PCBs by nonhuman primates. Since PCBs tend to be toxic in monkeys at levels lower than those that cause toxicity in rats, the authors suggested that arene oxide formation may play a larger role in PCB metabolism in monkeys than in rats.

Metabolism of Penta- and Hexa-chlorinated Isomers. In rats, penta-PCB isomers are metabolized similarly to di-, tri- and tetra-CBs. Goto et al. (1975) reported an administration of $[^{14}C]$-labeled penta-CBs to rats and recovery of urinary and fecal metabolites. Isomers given to rats were 2,4,6,2'6'-, 2,4,6,3'4'- or 2,4,6,3'5'-penta-CB. Details of protocol were not given. Fecal excretion of metabolites accounted for more of the radioactivity recovered than did urinary excretion. Urinary excretion occurred exclusively as conjugates, particularly glucuronides. Monohydroxylated metabolites predominated. Further analysis revealed that hydroxylation always occurred at a *meta*-position of the biphenyl ring. In the case of 2,4,6,2'6'-penta-CB, an additional metabolite (3',4'-dihydroxy-2,4,6,2',6'-penta-CB) was identified.

Chen et al. (1976) and Chen and Matthews (1974) reported on metabolites of 2,4,5,2',5'-penta-CB produced by male Sprague-Dawley rats. Treatment consisted of administration of 25 mg/kg bw daily for 5 days by gavage to five rats. Feces and urine were collected daily for 7 days. The major metabolite isolated was a monohydroxylated derivative found to be hydroxylated in the meta-position. A dihydroxy derivative was found as a minor metabolite. Urinary metabolites, which accounted for 6% of total radioactivity, were entirely in conjugated forms. Fecal metabolites, accounted for 62% of total radioactivity given. Further analysis revealed the presence of a 3,4-dihydrodiol as mentioned by Hsu et al. (1975a,b) for the metabolism of tetra-CB. These authors suggested an arene oxide intermediate as being the most probable mechanism resulting in the metabolites found.

As a part of the study described earlier, Goto et al. (1975) investigated the metabolism of 2,4,6,2',4',6'-HCB in rats. As they reported for the four penta-CB isomers tested, metabolism of this hexa-CB isomer yielded meta-monohydroxy analogs as the major metabolite. No other metabolites were mentioned. In the study by Hutzinger et al. (1972) described earlier, administration of 2,4,5,2',4',5'-HCB did not result in the formation of hydroxylated metabolites.

Sundstrom and coworkers (Sundstrom et al., 1976a,b; Jensen and Sundstrom, 1974) administered 2,4,5,2',4',5'-HCB to rats, mice and rabbits to study metabolism. The hexa-CB was disolved in peanut oil that was consumed freely by rats and mice. Rats consumed about 15 mg and mice an undetermined amount of the total compound. Rabbits were fed diets containing 1% of the test compound for 7 days, resulting in a total intake of

1 g/kg bw. Feces and urine were collected and extracted. Analysis of extracts from rats and mice yielded only one metabolite, the 3-hydroxy-derivative of HCB. Rabbits apparently metabolized this HCB somewhat differently, as other metabolites were identified as well. They included 3-hydroxy-2,5,2',4'5'-penta-CB and 4-hydroxy-2,5,2',4'5'-penta-CB that resulted from loss of a chlorine atom, and 4-hydroxy-2,3,5,2',4',5'-hexa-CB that resulted from a chlorine shift. The authors postulated a scheme of metabolism involving an arene oxide intermediate as the primary mechanism of metabolism. An earlier report by some of these authors (Hutzinger et al., 1974) presented the case for dechlorination of hexa-CB in the overall metabolic scheme for this compound.

Metabolic schemes have not been proposed for the more highly chlorinated biphenyls. Discussion of PCB metabolism organized around the metabolism of individual isomers tends to cause certain generalities to be overlooked. Kurachi (1983) and Kurachi and Mio (1983a), for example, reported that rats and mice exposed to di-, tetra- or penta-CBs excrete at least part of the metabolites as glutathione conjugates and other sulfur-containing compounds. The protocol of these studies was not reported in detail, but the PCBs to be tested were dissolved in salad oil and either injected i.p. or fed to rats and mice. In an excellent review, Safe (1980) summarized much of the current knowledge of PCB metabolism. Phenolic products are the major metabolites produced. Polyhydroxylated derivatives are also formed as are various methyl or methoxy derivatives. Trans-dihydrodriols are formed and may be a major PCB metabolite in certain species such as rabbits (Sundstrom et al., 1976a,b) and nonhuman primates (Hsu et al., 1975a,b). The finding of relatively large amounts of trans-dihydrodiols substantiated the argument for arene oxide intermediates. Arene oxides have been shown to be potentially electrophilic, and as such may cause alkylation of cellular macromolecules such as DNA, RNA and intracellular proteins. Metabolically mediated cytotoxicity, mutagenicity and carcinogenicity may result (Safe, 1980).

As the degree of chlorination increases, the rate of excretion, and probably metabolism, decreases. Apparently, the percentage of total dose that is excreted as unmetabolized compound is directly related to the degree of chlorine substitution at least up to the penta-isomer (Goto et al., 1974). Also related to degree of chlorine substitution seems to be the percentage of metabolites excreted in urine compared to fecal excretion. Metabolites of the more highly chlorinated isomers are excreted almost entirely through the feces (Goto et al., 1974).

Excretion

Rodent Studies. Yoshimura and Yamamoto (1975) suggested that PCBs may be excreted unchanged into the GI tract. A 1 mg dose of 2,4,3',4'-TCB was injected i.v. into each of 15 adult male Wistar King rats with or without ligated bile ducts. Contents of stomach, small intestine, cecum, large intestine and rectum were taken at 1, 3, 6, 18 and 24 hours postinjection. Fecal samples were collected daily for 4 days. Unmodified tetra-PCB was found in the feces at the end of each 24-hour period. Daily excretion ranged from 0.5-0.8% of total dose given. At the end of the 4-day collection period, excretion of unmodified compound accounted for 2.5% of the total dose. Unmodified tetra-CB was isolated from contents of small intestine as early as the postinjection (0.1% of original dose). During the period of 3 and 6 hours postinjection, 0.2 and 0.3%, respectively, of the original dose was recovered, unmodified, from contents of small intestine. Traces were also found in cecum and rectum. It was concluded that excretion of unmodified PCBs from the small intestine constituted a major mechanism of excretion in the rat. In all, however, fecal excretion of unmodified compound probably accounts for very little of the total excretion of compounds that are metabolized fairly extensively, which would include the less highly chlorinated PCBs.

Goto et al. (1974a,b,) injected radiolabeled 2,3-di-CB; 2,4,6-tri-CB; 2,3,5,6-TCB or 2,3,4,5,6-penta-CB into rats (see Metabolism Section). Collection and extraction of feces for 10 days was done to determine fecal excretion of unmodified parent PCB compounds. At the end of 10 days, fecal excretion of unchanged compound accounted for 5.7, 9.2, 7.0 and 23.2% of original dose of the di-, tri-, tetra- and penta-CBs, respectively. Broadly speaking, degree of fecal excretion of unchanged parent compound seemed to increase directly to the degree of chlorination or, presumably, inversely to the rate of metabolism.

Fecal and urinary excretion of metabolites of PCBs have been investigated in detail by several laboratories. Table III-6 contains summaries of half-lives from these studies. Chen and Matthews (1974), Matthews and Anderson (1975a,b), Chen et al. (1976), Tuey and Matthews (1977), Anderson et al. (1977) and Lutz et al. (1977) have intensively investigated the toxicokinetics of 4-MCB; 4,4'-DCB; 3,5,3'5'-TCB; 2,4,5,2'5'-penta-CB and 2,4,5,2'4'5'-HCB in the rat. Radiolabeled (^{14}C) isomers were injected i.v. in the amount of 0.6 mg/kg bw once into mature male Sprague-Dawley rats. There were three rats in each time group for each isomer tested.

TABLE III-6

Half-Lives of PCBs

Species/ Strain	Sex	Substance	Compartment	Half-life	Comments	Reference
Rat/Sprague-Dawley	M	2,4,5,2',4',5'-hexa-CB metabolite I	fecal	0.9 days	Metabolites were not characterized.	Muehlebach and Bickel, 1981
		metabolite II	fecal	8.8 days		
		metabolite III	fecal	100.5 days		
		metabolite I	urine	0.3 days		
		metabolite II	urine	7.4 days		
Mice/CBA	F	2,4,5,2',5'-penta-CB metabolites	fecal	6 days	All biliary metabolites combined	Berlin et al., 1975
Monkey/ rhesus	M,F	2,5,4'-tri-CB-4-biphenylol	urine	30-32 hours	Unable to characterize fecal metabolites	Felt et al., 1977
		4,2'5'-tri-CB-3-biphenylol conjugates	urine	4.5-4.8 days		
Mice/NR	F	2,3,2',3'-tetra	whole body	0.9 days	10 mg/kg each isomer in diet for 20 days	Mizutani et al., 1977
		2,4,2',4'-tetra	whole body	9.2 days		
			liver	7.8 days		
		2,5,2',5'-tetra	whole body	3.4 days		
			liver	2.6 days		
		3,4,3',4'-tetra	whole body	0.9 days		
		3,5,3',5'-tetra	whole body	2.1 days		
			liver	2.2 days		
		2,4,2',4'-tetra	whole body	4.9 days	Phenobarbital added 200 mg/kg diet: enzyme induction.	
		2,5,2',5'-tetra	whole body	2.6 days		
		3,5,3',5'-tetra	whole body	1.5 days		
Rat/Sprague-Dawley	M	4-mono- (1 metabolite)	fecal	15.7 hours	Metabolites were differentiated but not characterized	Matthews and Anderson, 1975b
		4,4'-di- (1 metabolite)	fecal	22.2 hours		
		2,4,5,2',5'-penta- (1 metabolite)	fecal	39.2		
		2,4,5,2',5'-penta- (another metabolite)	fecal	211 hours		
		2,4,5,2',4',5'-hexa- (1 metabolite)	fecal	49 hours		
		2,4,5,2',4',5'-hexa- (another metabolite)	fecal	642 hours		

Urinary excretion of total dose of injected radioactivity at the end of 24 hours was about 52, 24, 25 and <1% for the mono-, di-, tetra-, penta- and hexa-CB isomers, respectively. Urinary excretion was entirely in the form of conjugates of metabolites (see Metabolism Section). At the end of 24 hours, fecal excretion of four of the somers tested accounted for 20-30% of total dose of injected radioactivity. Fecal excretion of hexa-CB at the end of this time accounted for only about 2% of the total dose of injected radioactivity.

Half-lives of fecal excretion of single metabolites of these isomers were calculated to be 15.7 and 22.2 hours for metabolites of MCB and DCB, respectively. Half-lives of two separate metabolites of penta- and hexa-CB were found to be 39.2 and 211, and 49 and 642 hours, respectively (Matthews and Anderson, 1975b). The half-life of whole-body elimination of TCB was estimated, based on excretion, to be 9.75±3.44 days.

As expected, the half-life of fecal excretion of metabolites appeared to be directly related to degree of chlorination of the parent compound. The rate-limiting factors controlling excretion are probably those factors that control the rate of metabolism; with the exception of very low levels of metabolites in the blood and bile, tissue stores of PCBs are virtually entirely unmodified parent compound (Lutz et al., 1977). These authors therefore concluded that the rate of metabolism, hence excretion, is determined at least in part by the degree of chlorination of the parent PCB compound. The case of 3,5,3',5'-TCB is unique in that its half-life of excretion is in the same general range as that of the much more highly chlorinated HCB. This fact was thought to support the assumption that the presence of vicinal unsubstituted carbon atoms, particularly at the ortho-meta positions, facilitates the rate of metabolism of PCBs (Sundstrom, 1976a; Tuey and Matthews, 1977), and consequently, their excretion.

Muehlebach and Bickel (1981) reported in considerable detail the toxicodynamics of $[^{14}C]$-2,4,5,2'4'5'-HCB. They gave single i.v. doses of 0.6 or 3.6 mg hexa-CB/kg bw or oral doses of 0.6 mg/kg bw, once weekly for 6 weeks to groups of three male Sprague-Dawley rats. Fecal, urinary and tissue levels of hexa-CB and its metabolites were monitored for up to 40 weeks. They determined that urinary excretion of metabolites occurred rather quickly with half-lives of 0.3 and 7.4 days for each of two urinary metabolites. Fecal excretion consisted of three identified metabolites. Half-lives of these isomers ranged from 0.9-100.5 days. Urinary excretion accounted for only 7.4% of the total HCB dose and was essentially complete by 2 weeks. Fecal excretion at the end of 40 weeks accounted for 17.02% of total radioactivity administered. Metabolism of hexa-CB seemed to occur only from blood and liver tissue depots; adipose tissue seemed to form a nonmobile, nonmetabolizeable reservoir. It was suggested, on the basis of these studies, that a maximum of 18% of this hexa-CB isomer would ever be metabolized, regardless of dose or duration of exposure.

Mizitani et al. (1977) investigated the accumulation and subsequent elimination of several tetra-CB isomers from mice. Groups of 20 female mice were fed diets containing 10 mg of 2,3,2',3'-; 2,4,2',4'-; 2,5,2',5'-; 3,4,3',4'- or 3,5,3',5'-TCB/kg diet for 20 days. Mice were killed and examined for distribution of PCBs at various time intervals after 20 days of exposure. Biological half-lives for each isomer were calculated. The half-lives for whole body elimination of these tetra-isomers in mice were 0.9, 9.2, 3.4, 0.9 and 2.1 for 2,3,2',3'-; 2,4,2',4'-; 2,5,2',5'-; 3,4,3',4'- and 3,5,3'5'-, respectively. The authors noted substantial differences between the rates of elimination of these tetra-isomers but were unable to relate these differences to chlorine substitution patterns.

In a subsequent experiment, Mizitani et al. (1977) fed diets containing 200 mg phenobarbital/kg diet, as well as three of the tetra-CB isomers discussed above, to mice for 20 days. The addition of phenobarbitol, a known inducer of hepatic xenobiotic metabolizing enzymes, reduced the biological half-lives of the tested isomers considerably. Biological half-lives of 2,4,2',4'-; 2,5,2',5'- and 3,5,3',5'-TCBs were 4.5, 2.6 and 1.5 days, respectively, in phenobarbitol-induced mice.

In an earlier experiment Gage and Holm (1976) studied the influence of molecular structure on the excretion of 14 PCB isomers in mice. The protocol of this study has been discussed in the Distribution Section, and the isomers tested as well as 7- and 21-day concentrations in body fat are presented in Table III-3. Since the degree of storage in body fat is inversely related to the rate of metabolism, and hence excretion, rate of depletion from body fat may indicate relative rate of excretion from the body. It appeared that the 4,4'-; 3,3',4',6'-; 2,3,2',4',6'- and 2,3,4,2',4',6'-HCB isomers were eliminated most rapidly. These compounds had at least one pair of ortho-meta vicinal carbon atoms unsubstituted, a configuration thought to be important to a relatively rapid rate of metabolism and excretion. The hexa-isomers, 2,4,5,2',4',5'- and 2,3,4,2',4',5'-, appeared to be eliminated most slowly. It appeared that for hexa-CBs, one ortho-meta-vicinal unsubstituted pair of carbon atoms and chlorine substitutions at the 2' and 6' positions on the other phenyl ring are important for a relatively rapid rate of metabolism and elimination. No half-lives were calculated for these isomers.

In an earlier study, this laboratory (Berlin et al., 1975) investigated the toxicodynamics of 2,4,5,2',5'-penta-CB in mice. About 15 mg/kg bw was administered by intubation or by injection in the tail vein. Animals were killed at varying time intervals; up to 24 hours if administration had been by injection, or up to 32 days if administration had been by intubation. Feces were collected for metabolic studies. This compound appeared to have a half-life of about 6 days.

Studies in Monkey and Man. Felt et al. (1977) investigated the metabolism and elimination of $[^{14}C]$-2,5,4'-tri-CB in rhesus monkeys. A total of two male and two female monkeys were given 550 mg of tri-CB in fruit daily for 84 days. Urine and feces were collected in 24-hour aliquots. At the end of the 84-day exposure period, one monkey of each sex was killed, and a wide range of tissues was examined for presence of tri-CB and its metabolites. At the end of a 28-day recovery period, the other monkeys were similarly examined. Characterization of fecal metabolites was not successful. Total excretion seemed initially to be quite rapid but slowed after 5 days. Characterization of urinary metabolites revealed the presence of conjugates of 2,3-hydroxy- and 4-hydroxy-tri-CB. Half-lives of these metabolites were calculated to be 4.5-4.8 days and 20-32 hours, respectively. On the basis of total excreted and recovered radioactivity, the half-life of 2,5,4'-tri-CB in young rhesus monkeys appears to be 4.5-4.8 days.

In 1979 an outbreak of poisoning that was attributed to ingestion of rice thought to be contaminated with PCBs occurred in central Taiwan (Chen et al., 1982). Blood samples were taken from 17 patients of the ward and out-patient clinic of the Veterans General Hospital in Taipei. At least two, and often three, samples were taken from each patient 2-17 months apart. Samples were analyzed for specific PCB isomers so that relative rates of excretion could be ascertained. Those data are presented in Table III-7. Relative elimination rates of PCB isomers in humans seem to parallel the situation in animals. In general, the tetra- and penta-isomers tend to be eliminated more rapidly than other penta-, hexa- and septo-isomers. Calculated half-lives for the 2,4,5,2',4'-penta and 2,3,4,3',4'-penta isomers were 9.8 and 6.7 months, respectively, in blood.

Alteration of Excretion. The rate of excretion of PCBs or their metabolites may be modified by various biological factors. Administration of phenobarbital, a known hepatic enzyme inducer, for example, promoted the metabolism and excretion of tetra-CB isomers in mice. This study has already been discussed (Mizutani et al., 1977). Kurachi and Mio (1983b) investigated the effects of fat mobilization caused by fatigue, activated charcoal and the combination of the two on the rate of elimination of Kanechlor-400 or -500 from the tissues of ICR mice. Groups of 4-10 mice were fed diets containing about 100 mg of either Kanechlor/kg. Mice fed Kanechlor-400 for 20 days were subjected for 3 days to a rotary cage (8 rpm) to stimulate activity and cause fatigue. Mice were killed and their livers were analyzed for the presence of individual PCB isomers of Kanechlor-400 at 0, 1, 2 and 3 days of fatigue. The total content of liver PCBs increased substantially as fatigue time accumulated. Gas chromatographic examination

TABLE III-7

Comparative Rates of Elimination of Some Major PCBs from the Blood of PCB-Poisoned Patients[a]

Peak No.	No. of ClAtoms	Substitution Pattern	Elimination Rate[b]
9-4-a[c]	4	2,4,5,4'(?)	+ + +
9-4-b[c]	4	2,5,3',4'	+ + + + +
9-4-c[c]	4	2,4,3',4'	+ + + +
10-4[c]	4	2,3,3',4'(?)	+ + + + +
11-5-a[c]	5	2,5,2',3',5'	+ + + +
11-5-c[c]	5	2,4,2',4',5'	+ +
12-5-a[c]	5	2,3,2',4',5'	+ + + + +
12-5-b[c]	5	2,5,2',3',4'	+ + + +
12-5-c[c]	5	2,4,2',3',4'	+ + + +(?)
13-5-a[c]	5	2,3,6,3'4'	+ + + + +
13-5-b	5	---	+ + + + +
14-5	5	2,3,4,5,3'(?)	+ + + +
15-5-a[c]	5	2,4,5,3',4'	+ + +
15-5-b	5	----	+ +
16-5[c]	5	2,3,4,3',4'	+ + +
16-6[c]	6	2,4,5,2',4',5'	+
17-6	6	----	+ + + +
18-6[c]	6	2,3,4,2',4',5'	+
19-6[c]	6	2,3,4,2',3',4'	+ +
20-6[c]	6	2,3,4,5,3',4'	+
21-7[c]	7	2,3,4,5,2',4',5'	+
22-7[c]	7	2,3,4,5,2',3',4'	+

[a] Source: Chen et al., 1982

[b] + + + + + = rapid; + + + + = less rapid; + + + = slow; + + = very slow; + = extremely slow.

[c] Structural assignments of these PCBs are mainly based on a comparison of published data with the GC retention indices determined for the major PCBs in toxic rice-bran oil from Taichung (see text.)

showed a substantial increase in the levels of 2,4,5,2'5'-; 2,4,5,2',4',5'- and other highly chlorinated isomers which tend to accumulate in fat. Tri- and most tetra-isomers were not elevated as a result of fat mobilization. These authors concluded that those physiological factors that cause fat mobilization, such as fatigue from excessive activity, may predictably result in mobilization; hence, metabolism and excretion of those PCB isomers which tend to locate in body fat stores.

Subsequently, an experiment was conducted to determine the effect of activated charcoal on elimination of PCBs in three groups of moderately fatigued mice. The first group was kept 6 days in a rotary cage (2-3 rpm) and fed a diet containing about 100 mg/kg PCBs (unspecified) and 20 g/kg activated charcoal. The control to this group was subjected to the rotary cage for the same length of time, and fed a diet containing PCBs but no charcoal. Another group had been exposed to PCBs in the diet for 6 days previous to 6 days exposure to the rotary cage and a diet modified only with charcoal. The control to this group were subjected to PCBs in the diet for 6 days before exposure to the rotary cage and an entirely unmodified diet. Mice were killed at the end of the 6-day period in rotary cages, and liver, kidney and feces were analyzed for total PCB content that was reported as µg/animal.

Group one exposed to concurrent administration of PCBs and charcoal in the rotary cage had levels of 15.4, 0.2 and 92.1 µg in liver, kidney and feces, respectively. The control group subjected to the same regimen, sans charcoal, had PCB levels of 98.6, 2.8 and 27.2 µg, respectively, in the same body compartments. Clearly the concurrent addition of charcoal to the diet containing PCBs, decreased GI absorption of PCBs as evidenced by the markedly reduced liver and kidney levels of PCBs and increased level of fecal PCBs in charcoal-fed mice. The mice that received charcoal subsequent to 6 days exposure to PCBs had levels of 22.4, 0.2 and 88.3 μg PCBs in liver, kidney and feces, respectively. The control group, which did not receive charcoal in the diet subsequent to 6 days of PCBs in the diet, had levels of 102.2, 3.2 and 35.3 μg PCBs in the same body compartments, respectively. This was considered to be excellent evidence that activated charcoal had an "internal effect" on hastening excretion of PCBs when stress caused fat mobilization and consequent movement of PCBs between body compartments.

Finally, Kurachi and Mio (1983b) fed several organic substances in diets to mice which also contained about 100 mg/kg Kanechlor-400. Length of time was unspecified. Preliminary data suggested that unmetabolizeable sugars, such as arabinase and marmitol, and an acid hydrolysate of pectin may significantly reduce liver and kidney burdens of PCBs. Fukuba and Kanamori (1982) fed diets containing the polysaccharides konjak mannan, pectin, cundlan and dextran to male Donryu rats that were treated with Kanechlor-300 by gavage. Further details of experimental protocol were lacking. These workers concluded that fecal excretion was promoted and liver levels of PCBs were reduced by the feeding of pectin, either concurrently with or subsequent to, the administration of PCBs. Pectin appeared only to be effective at dietary levels of 50-100 g/kg diet.

Summary

Absorption of PCBs from the GI tract seems to be rather complete. Albro and Fishbein (1972) treated rats with 19 separate PCB isomers and determined that for each of them, absorption exceeded 90%. Allen et al. (1974a) achieved over 90% absorption of Aroclor 1248 from the diets of monkeys. Little information was available concerning quantitative absorption from dermal exposure. The only reported exception to this high level of absorption was by Norbach et al. (1978), in which 59-87% of TCB passed unchanged through the gastrointestinal tract. The fact that Miller (1944) caused mortality and lesions in guinea pigs by dermal application of PCBs comparable to those produced by oral administration, argues that there is dermal absorption.

Distribution of ingested or injected PCBs tend to follow a biphasic pattern. In rats, Matthews and Anderson (1975a) demonstrated with four pure PCB isomers that, following single oral or i.v. doses, initial distribution from the blood to muscle and liver tissue is fairly complete at the end of 1 day. Redistribution to fat, skin and other fat-containing organs then occurred and was fairly complete by 7 days. This laboratory (Matthews and Anderson, 1975a) and others (Muehlebach and Bickel, 1981; Van Miller et al., 1975; Berlin et al., 1975) determined that the more heavily chlorinated isomers redistributed more heavily to adipose tissue than did the more labile, less heavily chlorinated isomers. Other tissues that tended to accumulate PCBs were gonads, adrenals and kidneys (Kurachi and Mio, 1983b).

It has been shown that PCBs cross the placenta and locate in the fetus. Torok and Weber (1981) demonstrated a rather low transfer of PCBs to the fetal mouse through the placenta until the 17th day of gestation. This coincided with the time that the fetus consumed its amnionic fluid. Tissue levels in 18-day-old mouse fetuses were markedly higher than levels in younger fetuses. The work of Masuda et al. (1979) confirmed these findings and demonstrated further that tissue levels of offspring as a result of nursing were markedly higher than either fetal or dam tissue levels. They also noted that the lesser chlorinated isomers, being more rapidly metabolized, did not transfer through the milk as readily as those isomers which tend to accumulate in adipose. Curley et al. (1973) demonstrated placental and lactational transfer of PCBs in rats. They found whole body concentrations in 21-day-old weanlings, 5- to 10-fold the concentrations in 20-day-old fetuses. Mizunoya et al. (1974) indicated that tissue levels of PCBs in 28-day-old weaned rats exceeded tissue levels in their dams. Allen and Barsotti (1976) observed these same phenomena in fetal and nursing infant rhesus monkeys. Several studies (Miller, 1977; Kuwabara et al., 1979; Hofvander et al., 1981; Rogan et al., 1980; Wickizer et al., 1981) reported the concentrations of PCBs in breast milk of nursing women from various parts of the world. Reported levels ranged from 2.8 to 2.5 μg/mℓ.

Metabolism of the lesser chlorinated isomers generally occurs more rapidly than does metabolism of more heavily chlorinated isomers. Additionally, the position of chlorine substituents profoundly offsets rate and extent of metabolism. Metabolites produced from PCBs tend to be 3- or 4-hydroxy-products (Hass et al., 1979; Safe et al., 1975b). Glucuronide and other conjugates of these metabolites prevail in the urine (Block and Cornish, 1959; Safe et al., 1975a).

Chloride shifts occasionally occurred, particularly chlorine was shifted from ortho positions (Hass et al., 1977). Methoxy groups on the phenyl rings were also found, particularly in metabolites of tetra- and more heavily chlorinated isomers (Goto et al., 1974a,b). Van Miller et al. (1925) and Norback et al. (1976) postulated an arene oxide intermediate as a likely metabolic intermediate in the formation of these metabolites. The observation of trans-3,4-dyhydro-3,4-dihydroxy-metabolites in monkeys (Norback et al., 1976; Hsu et al., 1975a,b) and rabbits (Gardner et al., 1973) was interpreted as strong evidence for the presence of arene oxide formation.

Excretion of PCB metabolites is apparently exclusively in the conjugated form. Hass et al. (1977) mentioned glucuronides of hydroxylated metabolites in urine. Recently, Kurachi (1983) and Kurachi and Mio (1983a) reported that rats and mice excrete at least part of their urinary PCB metabolites as glutathione and other sulfur-containing conjugates.

In a excellent summary of PCB metabolism, Safe (1980) pointed out that metabolism seems to preferentially involve hydroxylation at the 4- or 3,4 positions. The presence of vicinal unsubstituted carbon atoms at the 3-4 positions may be helpful for, but not essential to, this process. As the degree of chlorination increases, rate of metabolism tends to decrease. Those PCBs that metabolize with more difficulty tend to be excreted almost exclusively through the biliary route, while metabolites of mono-, di- and tri-isomers may be eliminated through the urine nearly as much as through the feces.

Evidence has been presented that fecal excretion of PCBs in an unmetabolized form may occur. Yoshimura and Yamamoto (1975) recovered unmodified tetra-CB from the duodenal contents of rats injected intravenously with TCB. Daily excretion for 4 days ranged from 0.5-0.8% of the total dose/day. Goto et al. (1974a) determined that 4.7-23.2% of injected PCBs were excreted unchanged into the GI tract by the end of 10 days postinjection. In a comparison of the excretion of di-, tri-, tetra- or penta-isomers, they determined that the penta-isomer was excreted unchanged to the greater degree.

One laboratory (Chen and Matthews, 1974; Chen et al., 1976; Matthews and Anderson, 1975a,b; Tuey and Matthews, 1977; Anderson et al., 1977; Lutz et al., 1977) investigated in detail the metabolism and excretion of PCBs by the rat. They have determined that urinary excretion is entirely in the form of conjugates of metabolites. The degree of urinary excretion was found to be inversely related to the degree of chlorination or probably directly related to rate and degree of metabolism. Urinary metabolites accounted for 52% of the excretion of a mono-isomer and <1% of the excretion of a hexa-isomer.

Half-lives of different metabolites of a PCB isomer differ widely. Different metabolites of a hexa-isomer had half-lives of 49 and 642 hours. The rates of excretion are probably directly related to the rate of metabolism (Lutz et al., 1977), and therefore, to degree and position of chlorination. Mizutani et al. (1977) noted that feeding phenobarbitol, a known metabolic enzyme-inducer, in diets also containing PCBs reduced the half-

IV. HUMAN EXPOSURE

Humans may be exposed to chemicals such as polychlorinated biphenyls (PCBs) from a variety of sources, including drinking water, food, ambient air, occupational settings and consumer products. This analysis of human exposure to PCBs is limited to drinking water, food and ambient air because those media are considered to be sources common to all individuals. Even in limiting the analysis to these three sources, it must be recognized that individual exposure will vary widely based on many personal choices and on several factors over which there is little control. Where one lives, works and travels, what one eats, and physiologic characteristics related to age, sex and health status can all profoundly affect daily exposure and intake. Individuals living in the same neighborhood or even in the same household can experience vastly different exposure patterns. In the Exposure Estimation section of this chapter, available information is presented on the range of levels of human exposure and intake for PCBs from drinking water, food and ambient air for the 70 kg adult male. It is not possible to provide an estimate of the number of individuals experiencing specific combinations of intake from those three sources. However, the Summary section of this chapter provides some insight into the relative contributions of the three sources, especially drinking water, for the range of intake values suggested by the available data.

Exposure Estimation

Drinking Water. One Federal survey and state study were obtained which included PCBs as an analyte. The National Organic Monitoring Survey (NOMS) was conducted to determine the frequency of occurrence of specific organic chemicals (including PCBs) in finished water supplies of 113 cities nationwide (U.S. EPA, 1977). Data from three phases (referred to as NOMS I, NOMS II and NOMS III) of the study were collected over an 11-month period (March 1976 to January 1977) to reflect any long-term or seasonal variations. PCBs were not found in groundwater supplies sampled in NOMS I (minimum quantifiable limit = 0.12 μg/ℓ). Only a single finished groundwater sample in each of NOMS II and NOMS III contained detectable levels of PCBs (~6% frequency of occurrence for both phases). Concentrations of 0.1 μg/ℓ were reported for the NOMS II and NOMS III samples (minimum quantifiable limits ranged from 0.1-0.2 μg/ℓ, respectively). PCBs were detected in finished surface water supplies in all three phases of NOMS. In NOMS I, concentrations of two samples observed were 0.13 and 1.4 μg/ℓ (mean = 0.77 μg/ℓ, frequency of occurrence = 2.2%). For NOMS II, two samples contained 0.1 μg/ℓ and one sample had a level of 0.2 μg/ℓ (mean = 0.13 μg/ℓ, frequency of occurrence = 3.3%). Only a single sample in NOMS III contained PCBs at a level of 0.2 μg/ℓ (frequency of occurrence = 1.1%). During a groundwater study in the state of New Jersey, Tucker and Burke (1978) examined 163 wells in all nine counties of the state, including public and private drinking water wells and wells near industrial sites and landfills. Fifty organic chemicals, pesticides and metals were analyzed for, including PCBs. Thirty-two of the 163 wells were positive for PCBs with concentrations ranging from 0.06-1.27 μg/ℓ. (The minimum reportable concentration was 0.06 μg/ℓ.) The highest concentration reported, 1.27 μg/ℓ, was from a well in Mercer County, NJ. It should be noted that many of the wells sampled were from highly populated, industrialized areas. The levels of PCBs observed in drinking water, when they have been found, are in the very low μg/ℓ range. Assuming that

PCBs are present at the highest concentration reported, i.e., 1.4 μg/ℓ in the NOMS I survey, the daily intake for a 70 kg adult male consuming 2 ℓ of drinking water per day would be 2.8 μg/day or 0.04 μg/kg/day.

Diet. Limited information was obtained on dietary exposure to PCBs. Dietary PCB intake has been reported for adults, as well as infants and toddlers, in the Food and Drug Administration's (FDA) Market Basket Studies for fiscal year 1979 (FDA, 1982a,b). The relative daily intakes of PCBs are presented for FY74 through FY79 in Table IV-1. There is no apparent trend in intake levels over the years for which information was obtained. Total dietary intake of PCBs for adults in FY79 was 0.0133 μg/kg/day (FDA, 1982a). [This includes an estimated 0.0053 μg/kg/day from dairy products, 0.0075 μg/kg/day from meat, fish and poultry, and 0.0005 μg/kg/day from oils and fats.]

No PCBs were detected for infant and toddler dietary studies by FDA in FY79 (FDA, 1982b). Total dietary intakes of PCBs for infants and toddlers in FY78 (as reported in FDA, 1982b) were 0.0113 and 0.0985 μg/kg/day, respectively. Information on the individual food groups comprising the total intake values was not obtained. The FDA calculations assume that the average infant weighs 8.2 kg (6-month-old) and the average toddler (2-year-old) weighs 13.7 kg. No comparisons of intakes of PCBs by geographic region were presented in the FDA Market Basket Studies.

Air. No information was found on inhalation exposure to PCBs. PCBs usually exhibit low vapor pressures and, thus, tend not to be found in ambient air (Halper, 1984). It is, therefore, assumed that ambient air does not contribute to human exposure to PCBs.

TABLE IV-1

Estimated Dietary Intake of PCBs for Adults, Infants and Toddlers[a] (μg/kg/day)

	Adult	Infant	Toddler
FY79	0.0133	ND	ND
FY78	0.0269	0.0113	0.0985
FY77	0.0164	0.0253	0.0301
FY76	T[b]	T	ND
FY75	T[b]	T	T
FY74	0.0056	--	--

[a] Source: FDA, 1982a,b

[b] In the FDA report that lists the intakes (FDA 1980a), values for the adult in FY75 and FY76 were reported as 0.0000 μg/kg/day. These values were given to indicate that only trace amounts of PCBs were detected in the market basket studies.

ND = Not detected

T = Trace

-- = Data not obtained

Summary

Table IV-2 presents a general view of the total amount of PCBs potentially received by a 70 kg adult male from air, food and drinking water. Seven separate exposure levels for drinking water and three levels for food (representing a probable range of exposure levels based on the data presented in the Exposure Estimation section) are shown in the table. It is assumed that air exposure is insignificant. The data presented have been selected from an infinite number of possible combinations of concentrations for the three sources. Whether exposure occurs at any specific combination of levels is not known; nor is it possible to determine the number of persons that would be exposed to PCBs at any of the combined exposure levels. The data presented represent possible exposures based on the occurrence data and the estimated intakes. The values presented in Table IV-2 for food levels of PCBs, as well as the values for drinking water levels, represent a range from the values found in the PCB monitoring data (see Drinking Water and Diet Sections). The intake from ambient air is assumed to be none. Assuming the intermediate food intake of 0.01 μg/kg/day and the intake of 0 μg/kg/day from air to be representative, drinking water would be the predominant source of PCB exposure in the adult male when drinking water levels exceed 0.35 μg/ℓ. An accurate assessment of the number of individuals for which drinking water is the predominant source of exposure cannot be determined from the data since specific locations containing high concentrations of PCBs in drinking water and low concentrations of PCBs in ambient air and food are unknown.

TABLE IV-2

Estimated Intake of PCBs from the Environment by Adult Males

Concentration in Drinking Water (μg/ℓ)	Estimated Total Intake in μg/kg/day (% from drinking water) Based on Food Levels of: Low (0.005 μg/kg/day)	Intermediate (0.01 μg/kg/day)	High (0.03 μg/kg/day)
0.0	0.005 (0%)	0.010 (0%)	0.030 (0%)
0.05	0.006 (22%)	0.011 (12%)	0.031 (4%)
0.1	0.008 (37%)	0.013 (22%)	0.033 (9%)
0.2	0.011 (53%)	0.016 (36%)	0.036 (16%)
0.5	0.019 (74%)	0.024 (58%)	0.044 (32%)
1.0	0.034 (85%)	0.039 (74%)	0.059 (49%)
1.4	0.045 (89%)	0.05 (80%)	0.07 (57%)

Intake from each source:

Water:	0.05 μg/ℓ:	0.0014 μg/kg/day
	0.1 μg/ℓ:	0.0029 μg/kg/day
	0.2 μg/ℓ:	0.0057 μg/kg/day
	0.5 μg/ℓ:	0.014 μg/kg/day
	1.0 μg/ℓ:	0.029 μg/kg/day
	1.4 μg/ℓ:	0.04 μg/kg/day

Air: 0 μg/kg/day

Food: 0.005 μg/kg/day
0.01 μg/kg/day
0.03 μg/kg/day

The relative source contribution data are based on estimated intake and do not account for a possible differential absorption rate for PCBs by route of exposure. In addition, the effects of the chemical on the body may vary by different routes of exposure.

REFERENCES

FDA (Food and Drug Administration). 1980a. Compliance program report of findings. FY77 total diet studies -- Adult (7320.73). Food and Drug Administration, U.S. Department of Health, Education and Welfare, Washington, DC.

FDA (Food and Drug Administration). 1980b. Compliance program report of findings. FY77 total diet studies -- Infants and Toddlers (7320.74). Food and Drug Administration, U.S. Department of Health, Education and Welfare, Washington, DC.

FDA (Food and Drug Administration). 1982a. Compliance program report of findings. FY79 total diet studies -- Adult (7320.73). Food and Drug Administration, U.S. Department of Health and Human Services, Washington, DC. FDA/BF-82/98.

FDA (Food and Drug Administration). 1982b. Compliance program report of findings. FY79 total diet studies -- Infant and toddlers (7305.002). Food and Drug Administration, U.S. Department of Health and Human Services, Washington, DC. FDA/BF-82/97.

Halper, M. 1984. Personal communication between Marty Halper, Director, Exposure Evaluation Division, Office of Pesticides and Toxic Substances, U.S. Environmental Protection Agency and Denis Borum, JRB Associates. September 5, 1984.

Tucker, R.K. and T.A. Burke. 1978. A second preliminary report on the findings of the state groundwater monitoring project. December, 1978. Program on environmental cancer and toxic substances. Department of Environmental Protection, State of New Jersey.

U.S. EPA. 1977. National Organic Monitoring Survey. Computer data file provided by the Technical Support Division, Office of Drinking Water, U.S. Environmental Protection Agency, Cincinnati, OH.

V. HEALTH EFFECTS IN ANIMALS

General Toxicity

Acute and Short-Term Toxicity. Acute and short-term toxicity information has been generated by many workers in many species. Several different products have been tested, each containing different proportions of PCB isomers and contaminants. Studies of up to 90 days of exposure are included in this Section. LD_{50} data are summarized in Table V-1 and discussed in the Toxicity Values Section. Multiple exposure studies are summarized by species and route of exposure in Tables V-2 to V-5, and discussed in the sections on acute and short-term toxicity in the rat, mouse and other species. Most of the acute toxicity studies have been conducted with rats, and data for this species are discussed first; toxicity in other species is discussed subsequently.

Toxicity Values -- LD_{50} values for several commercial mixtures and purified isomers are presented in Table V-1. Several reviews (Peakall, 1972; Nelson et al., 1972; Fishbein, 1974; Kimbrough, 1974; NIOSH, 1977; Kimbrough et al., 1978; McConnell, 1980) presented both LD_{50} data and a further discussion of the acute toxic effects of PCBs. Single-dose oral LD_{50}s in the rat ranged from 1.01 g Aroclor 1254/kg bw (Garthoff et al., 1981) to 3.98 g Aroclor 1221/kg bw (Fishbein, 1974). Mice appeared to be more sensitive, with single-dose oral LD_{50}s ranging from 19 mg of 3,4,5,3',4',5'-HCB/kg bw (Biocca et al., 1981) to 7.86 g of 2,4-DCB/kg bw (Kimbrough et al., 1978). Guinea pigs appeared to be the most sensitive species studied, with a reported oral LD_{50} of 0.5 mg of 3,4,5,3',4',5'-HCB/kg bw (McConnell, 1980).

TABLE V-1

Lethal Doses of PCBs

Species/ Strain	Sex/No.	Source of PCB	Route	LD_{50}	Comments	Reference
Guinea pig/ Hartley	NR	3,4,5-sym. HCB	oral	1.39 µmol/kg bw (0.5 mg/kg)	Single dose Half of the animals died within 30 days (LD_{50-30})	McConnell and Kinney, 1978
Guinea pig/ NR	NR/8	NR; ~42% Cl	oral	NR	Two 69 mg doses 7 days apart Mortality 11-29 days; centrilobular liver necrosis and fatty infiltration	Miller, 1944
Rat/Wistar	M-F/6	Aroclor 1254	oral	1.3 g/kg bw (30 days old)	Single doses Immature seemed more susceptible than older rats; sex did not seem to to be a factor; majority died within 3 days	Grant and Phillips, 1974
	M-F/6			1.4 g/kg bw (60 days old)		
	M/6			2.0 g/kg bw (120 days old)		
	F/6			2.5 g/kg bw (120 days old)		
Rat/Sherman	F/10	Aroclor 1254	i.v.	358 mg/kg bw	Adult; single dose; death in 5-110 min; dyspnea, depression, salivation, diarrhea	Linder et al., 1974
	NR/10	Aroclor 1254 Aroclor 1260	oral	4-10 g/kg	Adults, single dose	
	M/10	Aroclor 1254		1.295 g/kg	Weanlings, single dose: diarrhea, depression, salivation, death 1-3 days	
	M/10	Aroclor 1260	oral	1.315 g/kg	Weanlings: diarrhea, depression, salivation, death 1-7 days	
Rat/Sprague-Dawley	M/NR	Aroclor 1242		4.25 g/kg	14-day LD_{50}	Bruckner et al., 1973; Kimbrough et al., 1978
Rat/Wistar	M/NR	Kanechlor-400	oral	1.30 mL/kg		Kimbrough et al., 1978
	F/NR	Kanechlor-400	oral	1.14 mL/kg		
	M/NR	Kanechlor-300	oral	1.15 g/kg		
	F/NR	Kanechlor-300	oral	1.05 g/kg		

TABLE V-1 (cont.)

Species/ Strain	Sex/No.	Source of PCB	Route	LD_{50}	Comments	Reference
Rat/NR	NR	Aroclor 1221	oral	3.98 g/kg	Toxicity apparently decreasing with increasing chlorine substitution	Fishbein, 1974; Nelson et al., 1972
		Aroclor 1232	oral	4.47 g/kg		
		Aroclor 1242	oral	8.65 g/kg		
		Aroclor 1248	oral	11 g/kg		
		Aroclor 1260	oral	10 g/kg		
		Aroclor 1262	oral	11.3 g/kg		
		Aroclor 1268	oral	10.9 g/kg		
		Aroclor 4465	oral	16 g/kg		
		Aroclor 5442	oral	10.6 g/kg		
		Aroclor 5460	oral	19.2 g/kg		
		Aroclor 2565	oral	6.31 g/kg		
Rat/Sherman	F/NR	Aroclor 1221	oral	4.0 g/kg		Nelson et al., 1972
		Aroclor 1262	oral	11.3 g/kg		
Rat/Osborne-Mendel	M/NR	Aroclor 1254	oral	1.01 g/kg	Single observation period	Garthoff et al., 1981
	M/NR	Aroclor 1254	oral	1.53 g/kg	5 multiple doses 2 times/week	
	M/NR	Aroclor 1254	oral	1.99 g/kg	5 multiple doses 1 time/week	
Rabbit/NR		Aroclor 1221	dermal	2.000-3.169 g/kg		Nelson et al., 1972
		Aroclor 1232	dermal	1.26-2.0 g/kg		
		Aroclor 1242	dermal	0.794-1.269 g/kg		
		Aroclor 1248	dermal	0.794-1.269 g/kg		
		Aroclor 1260	dermal	1.26-2.0 g/kg		
		Aroclor 1262	dermal	1.26-3.16 g/kg		
		Aroclor 1268	dermal	2.5 g/kg		
Mice/CFI	M/NR	Kanechlor 400	oral	1.875 mℓ/kg		Kimbrough et al., 1978
Mice/CFI	F/NR	Kanechlor 400	oral	1.57 mℓ/kg		
Mice/dd	F/NR	2',4'-dichlorobiphenyl	oral	7.86 g/kg	Data reviewed suggest increased toxicity in mice with greater	Kimbrough et al., 1978
Mice/DVI	NR	trichlorobiphenyl	oral	3.06-4.25 g/kg	chlorine substitution	
Mice/DVI	NR	2,4,3',4'-tetrachlorobiphenyl	i.p.	2.15 g/kg		
Mice/CFI	NR	2,3,4,3',4'-pentochlorobiphenyl	i.p.	0.65 g/kg		

TABLE V-1 (cont.)

Species/ Strain	Sex/No.	Source of PCB	Route	LD_{50}	Comments	Reference
Mice/CBA	NR	Aroclor 1254	i.p.	880 mg/kg	5-day observation period	Lewis et al., 1972
Mice/ALAS	NR	Aroclor 1254	i.p.	1200 mg/kg	5-day observation period	
Mice/BALB/CJ	NR	Aroclor 1254	i.p.	1080 mg/kg	5-day observation period	
Mice/CSF	NR	Aroclor 1254	i.p.	1000 mg/kg	5-day observation period	
_Peromysens maniculatus_*	NR	Aroclor 1254	i.p.	970 mg/kg	5-day observation period	
Mice/C57Bl/6J	M/5	3,4,5-Sym. HCB 2,4,6-Sym. HCB 2,4,5-Sym. HCB 2,3,6-Sym. HCB	oral oral oral oral	19 mg/kg 21.4-64.3 mg/kg >64.3 mg/kg >64.3 mg/kg	28-day LD_{50} 28-day LD_{50} 28-day LD_{50} 28-day LD_{50}	Biocca et al., 1981
Rabbits	NR	NR	NR	8-11 g/kg		Clegg, 1971; Penkall, 1975
Mink/Pastel	NR/81	Aroclor 1221, 1242, 1254	gavage	500-4000 mg/kg	Single doses	Aulerich and Ringer, 1977

*Field mouse

NR = Not reported

TABLE V-2

Acute and Short-Term Toxicity of Orally-Administered PCBs to Rats

Strain	Sex/No.	Weight/Age	Source of PCBs/Vehicle	Dose/Duration	Animal Effects	References
Sprague-Dawley	F/56	200-300 g	Aroclor 1254/diet	0-900 mg/kg diet for 10 days	≥300 mg/kg: weight loss. Decreased placental protein, glycogen; partial anorexia.	Spencer, 1982
Sprague-Dawley	M/18	250-300 g	Aroclor 1254/corn oil	50 mg/kg bw for 7 days	Increased acid phosphatase in testicular interstitial cells.	Dikshith et al., 1975
Holtzman	F/25	sexually mature	Aroclor 1254/peanut oil	0-64 mg/kg day for 7 days	64 mg/kg/day: increased liver weight.	Sager, 1983
Fischer	M/42	weanlings	Aroclor 1254/diet	0, 71, 179, 357, 429, 700, 1400 mg/kg diet for 14 days	Depressed rate of gain, food intake; increased liver weight.	Carter and Mercer, 1983
Sprague-Dawley	M/12-18	200-250 g/ 7 weeks	Aroclor 1254/mineral oil	0, 0.05, 0.25, 0.5 g/kg bw daily for 21 days	≥0.05 g/kg bw/day: decreased rate of gain to frank weight loss; depressed feed intake; depressed water intake; depressed body temperature.	Komives, 1979; Komives and Alayoku, 1980
Wistar	F/40	C1/1 year	Aroclor 1254/corn oil	0, 12.5, 25.0, 50, 100, 400, 800 mg/kg bw daily for 7 days	≥400 mg/kg: mortality; ≥100 mg/kg: increased liver fat percent ($p<0.05$). All doses: increased liver weight ($p<0.05$).	Grant and Phillips, 1974
Wistar	M-F/144	30, 60 or 120 days	Aroclor 1254/corn oil	0, 5, 10, 20 mg/kg/ day for 7 days	All doses: increased liver weights, increased aniline hydroxylase activity.	Grant and Phillips, 1974
Long-Evans	M/125, F/25	5 weeks	2,5,2',5'-tetra-CB/ corn oil	0, 0.5, 1.0, 1.5, 2.0 g/kg bw, single dose	≥1.0 g/kg: heavy mortality; thymic hemorrhage; atrophy; liver and kidney enlargement; splenic and lymph node regression.	Allen et al., 1975
Hooded	M/NR	180-200 g	Aroclor 1254/diet	250 mg/kg diet for 14 days	Increased thyroid activity.	Bastomsky, 1974
Sprague-Dawley	F/6	200-250 g	^{3}H-labelled 2,5,2',5'-TCB/ corn oil	0, 1.7 g/kg bw, single dose	Intracellular vacuolization; vesiculation, fragmentation of rough ER, enlarged, varified cytoplasm; altered mitochondrial conformation; ATP-ase inhibition.	Lin et al., 1979
Sprague-Dawley	F/24, M/24	300 g	Phenoclor DP6/diet	0, 100 mg/kg diet for 8 days	Decreased phenobarbital sleeping time, increased liver weight and protein content; males more greatly affected than females.	Narbonne, 1979

TABLE V-2 (cont.)

Strain	Sex/No.	Weight/Age	Source of PCBs/Vehicle	Dose/Duration	Animal Effects	References
Sprague-Dawley	M,F/48	adult and 35 days	Phenoclor DP6/diet	13 mg/kg diet adult 24 mg/kg diet young for 8 days	Liver weight elevated equally in both sexes; elevated liver protein and fat more noticeable in adults.	Narbonne, 1979
Fischer	M/50	34 days	Aroclor 1254/diet	0, 20 mg/kg diet for 1, 2, 4, 8, or 14 days	Hepatomegaly by day 4.	Carter, 1983
CD	F/3/ group		4-monochlorobiphenyl/ cottonseed oil	30 mg/kg bw/day on days 8, 11, 13, 15, 18 of pregnancy	Elevated intestinal monoamine oxidase, serum sorbitol dehydrogenase and alkaline phosphatase.	Wolden et al., 1982
Long-Evans	M/12	130 g	Aroclor 1248/diet	0 or 100 mg/kg diet for 4 weeks	Hepatomegaly (4.46% of bw), obvious increase in SER.	Allen et al., 1975
			2,5,2',5'-tetra-CB		Hepatomegaly (3.38% of bw) less obvious increase in SER.	
Wistar	M,F/38	NR	Aroclor 1254/diet	1000 mg/kg diet for 14 or 30 days	At ≥14 days, 30-72% reduction in rate of gain; at 30 days, 29% reduction in food intake; no change in liver weight. Altered cholesterol, fatty acid synthesis.	Kling et al., 1978
Osborne-Mendel	M/6/ group	8 weeks	Aroclor 1254/diet	0, 5, 50, 500 mg/kg diet for 4 weeks	≥5 mg/kg diet: enlarged thyroid, reduced follicle size, follicular lumen reduced, papillary projections and cytoplasmic projections, diluted RER.	Collins and Capen, 1980b
Gunn	M/12/ group	300-400 g	Aroclor 1254/diet	500 mg/kg diet for 42 days	Thyroid follicular cells more columnar, diluted RER, vacuolated mitochondria.	Collins and Capen, 1980a
Osborne-Mendel	M/NR	8 weeks	Aroclor 1254/diet	0, 50, 500 mg/kg diet for 4 or 12 weeks	≥50 mg/kg diet, by 4 weeks: enlarged thyroid, follicular cells more columnar, vacuoled cytoplasm, papillary projections, cytoplasmic processes. Dilated RER, Golgi apparatus more prominent, larger number of enlarged lysosomes. Reduced serum thyroxin; after 35-week recovery period thyroids resemble those of controls, serum thyroxin normal.	Collins et al., 1977

TABLE V-2 (cont.)

Strain	Sex/No.	Weight/Age	Source of PCBs/Vehicle	Dose/Duration	Animal Effects	References
Sprague-Dawley	M/40	100-130 g	Kanechlor 500/diet	100 mg/kg diet for 4 weeks	No effect on food intake, water intake, urinary volume. Slightly enlarged brain, spleen and liver. Hematologic values unchanged. Serum protein, cholesterol, cholinesterase elevated; serum triglyceride reduced.	Oishi et al., 1978
Holtzman	M/6-16/groups	ca 250 g	Aroclor 1254/diet	0, 5, 50 or 500 mg/kg diet for 2, 3 or 5 weeks	500 mg/kg: weight loss. ≥5 mg/kg: reduced food intake, hepatomegaly. 500 mg/kg: slightly increased size of kidney, testes; decreased adiposa. 50-500 mg/kg: liver - increased content of fat; decreased protein, RNA, DNA. 50-500 mg/kg: blood - glucose reduced, BUN, cholesterol, protein increased. 5 mg/kg: aminopyrine dimethylase activity increased. 50-500 mg/kg: p-nitrobenzoate reductase, pentobarbital hydroxylase activity increased.	Garthoff et al., 1977
Sprague-Dawley	M/10/group	weanlings	Aroclor 1254 or 1260/diet	0 or 20 mg/kg diet for 28 days	No effect on food intake, weight gain or liver size. Moderate liver lobular pattern with periportal perinuclear halos and perivenous cytoplasmic ballooning; anisokaryosis, moderate fatty liver degenerations. Thyroid changes (see Collins et al., 1977). Reduced SGOT; induced mixed function oxidases.	Chu et al., 1977
Wistar/Neuherberg	M/10/group	100 g	Clophen A-50/olive oil	0, 2, 10, 50, 150 or 250 mg/kg bw, twice weekly for 6 weeks	≥50 mg/kg: decreased bw; slightly increased food intake; increased liver weight. ≥150 mg/kg: elevated SGOT, SGPT. ≥50 mg/kg: increased serum bilirubin, protein, triglycerides; increased urinary porphyrin precursors. >2 mg/kg: increased serum cholesterol; hepatic coagulation focal necrosis, PMN infiltration, fatty degeneration, enlarged (functional) nucleoli.	Baumann et al., 1983
NR	M/10	6 months	"PCBs-1221"/ethyl alcohol	250 mg/ℓ in drinking water for 10 weeks	Elevated plasma corticosterone; hyperactivity of zona fasciculata of adrenal cortex.	Wasserman et al., 1973

TABLE V-2 (cont.)

Strain	Sex/No.	Weight/Age	Source of PCBs/Vehicle	Dose/Duration	Animal Effects	References
Sprague-Dawley	M/NR	100 g	Aroclor 1248, 1254 or 1262/diet	0 or 1000 mg/kg diet up to 6 weeks	Reduced rate of gain: Aroclor 1248 > 1254 > 1262. Moderate elevation of Hb, PCV. Relative neutrophilia, lymphocytopenia. Enlarged liver, decreased thymus, fatty liver degenerations, cystic areas and focal necrosis with infiltration of inflammatory cells. ER proliferation, vesiculation of RER, increased number of lysozymes. Increased hepatic protein, RNA, phospholipid; decreased DNA, cholesterol. Induction of N-dimethylase, nitroreductase.	Allen and Abrahamson, 1973
Wistar	F/8	112-130 g	Phenoclor DO6/diet	2000 mg/kg diet up to 56 days	>3 weeks, UV fluorescence of incisors, small intestine (porphyria). Hepatic enlargement with centrilobular degeneration. Splenic degeneration with disappearance of white pulp reduction in red pulp, evidence of siderosis.	Vos and Koeman, 1970
Sprague-Dawley	M,F/4/group	NR	Clophen A-50/olive oil	0 or 100 mg/kg bw 1 time/week for 7 weeks	M: increased liver percent of bw from 2.6-3.3%. Presence of ATPase deficient islands. F: increased liver percent of bw from 2.9-3.6%. Greater presence of ATPase deficient islands.	Deml and Oesterle, 1982
Holtzman	M/30	NR	Aroclor 1254/diet	0, 5, 50, 500 mg/kg diet for 5 weeks	$\geq$50 mg/kg: hepatomegaly, fatty degeneration, hepatocellular hypertrophy and cytoplasmic vacuolization. $\geq$5 mg/kg: enlarged SER; decreased number of mitochondria, lysosomes; increased Golgi apparatus. $\geq$50 mg/kg: Golgi apparatus decreased.	Kasza et al., 1978b
Holtzman	M/30	NR	Aroclor 1254/diet	0, 5, 50 or 500 mg/kg diet for 5 weeks	$\geq$5 mg/kg: enlarged thyroid, reduction in follicular size, hyperplastic cells with papillae and cytoplasmic processes extending into lumenal colloid; Follicular cells more columnar, mitochondria vacuolated with disrupted cristae, accumulation of colloid, increased number of lysosomes.	Kasza et al., 1978a

NR = Not reported

TABLE V-3

Acute and Short-Term Toxicity of Orally-Administered PCBs to Mice

Strain	Sex/No.	Weight/Age	Source of PCBs	Vehicle	Dose/Duration	Animal Effects	Reference
NMRI	F/116	Sexually mature	2,4',5-tetrachlorobiphenyl	Peanut oil	0, 0.05, 0.5 mg/day	0.50 mg/day: increased cytochrome P-450	Orberg, 1978
NMRI	F/108	Sexually mature	2,2',4,4',5,5'-hexachlorobiphenyl	Peanut oil	0, 0.05, 0.5 mg/day for 6 days	≥0.05 mg/day: increased cytochrome P-450 (PL0.005). 0.5 mg/day: hepatomegaly	
CDI	F/NR	60-90 days	3,3',4,4'5,5'-hexachlorobiphenyl	Cottonseed oil	0-16 mg/kg/day for 10 days	≥8 mg/kg/day: decreased weight gain, lethargy, vaginal bleeding of pregnant females	Marks et al., 1981
CBH	F/10/group	12 weeks	2,2',4,4',5,5'-hexa-CB	Peanut oil	0, 0.5 mg/day for 7 or 13 days	Increased liver weights; reduced sensitivity to stressful stimuli (moving)	Mattsson et al., 1981
ICR	M/5/group	Adult	Aroclor 1254	Diet	0, 62.5, 250, 1000 or 4000 mg/kg diet 14 days	4000 ppm: total mortality by day 7. 1000 ppm: death of 3/5 by day 15. 250 ppm: hepatomegaly, depressed food intake, decreased pentobarbital sleeping time. 62.5 ppm: elevated serum corticosterone.	Sanders et al., 1974
CD-1	M/9/group	25-29 g	Aroclor 1254	NR	500 mg/kg, single dose	Prolonged pentobarbital-induced sleep time.	Rosin and Martin, 1983

TABLE V-3 (cont.)

Strain	Sex/No.	Weight/Age	Source of PCBs	Vehicle	Dose/Duration	Animal Effects	Reference
C57Bl/6J	M/5/group	18-20 g, 5 weeks	3,4,5-sym. HCB... 2,4,6-sym. HCB... 2,4,5-sym. HCB... 2,3,6-sym. HCB...	Diet	0, 0.3, 1, 3, 10 or 30 mg/kg diet for 28 days 0, 10, 30, 100 or 300 mg/kg diet for 28 days	Lowest dose to cause: Enlarged Liver / Reduced Spleen Thymus / Increased Testis Weight ...0.3 / 10 / 10 ...100 / 100 / NR ... 30 / 300 / NR ... NR / NR / NR 300 mg/kg Diet / Levels in Adipose / Levels in Liver 3,4,5- / 6912 / 1344 2,4,6- / 4329 / 1022 2,4,5- / 3923 / 637 2,3,6- / 280 / 52 30 mg/kg 3,4,5-HCB depressed serum protein, caused UV fluorescence of liver, teeth, sternum. Liver: 1.0 mg/kg 3,4,5-HCB caused liver microabscesses to severe fatty degeneration and necrosis at 30 mg/kg. Other isomers: same lesions at 300 mg/kg. Thymus: 3.0 mg/kg 3,4,5-HCB caused moderate to marked involution at 30 mg/kg; 300 mg/kg 2,4,6-HCB caused marked involution; 300 mg/kg 2,3,6-HCB caused slight involution. Spleen: 3.0-30 mg/kg 3,4,5-HCB, 300 mg/kg 2,4,6-HCB caused moderate depletion of lymphocytes. 3,4,5-HCB: enlarged spermatagonis. 2,4,6-HCB: cardiomyopathy and passive congestion of liver, lung.	Biocca et al., 1981
Swiss Albino	M/10/group	23-47 g	2,4,5,2',4',5'-HCB	Peanut oil	0, 200, 500, 1000 mg/kg bw/day for 28 days	No significant effect on body weight, food or water intake,fecal or urinary output. 1000 mg/kg: decreased kidney weight; 500 mg/kg: increased liver weight. Brain, testes, spleen; no change. PCV: no change. Hepatocytes: enlarged cytoplasm and nuclei; fatty infiltration, reduced glycogen, increased SER, 2; lysosomes, mitochondria, RER. No deficits in neuromuscular coordination.	Carter and Cameron, 1977

TABLE V-3 (cont.)

Strain	Sex/No.	Weight/Age	Source of PCBs	Vehicle	Dose/Duration	Animal Effects	Reference
BALB/CJ	M/-13/group	18-20 g	Aroclor 1242	Diet	0 or 167 mg/kg diet for 6 weeks	Increased (p<0.05) mortality caused by Salmonella typhosa endotoxin at 6 weeks. (Hepatocytic.) Increased (p<0.05) mortality caused by Plasmodium berghei at 3 weeks. Hypertaophy; no histological (Hepatocytic.) alteration of lymphoid tissues.	Loose et al., 1978b
BALB/CJ	M/-15/group	18-20 g	Aroclor 1242	Diet	167 mg/kg diet for 6 weeks	>2-fold reduction in primary and secondary splenic plaque-forming cells in response to sheep RBC, decreased serum immunoglobulin. Decreased memory cell function and serum IgA when immunized with sheep RBC 3 weeks before 6-week exposure to Aroclor 1242.	Loose et al., 1978a
			Aroclor 1016 or 1242	Diet	167 mg/kg diet for 6 weeks	Increased (p<0.05) mortality caused by S. typhosa endotoxin. (No difference between Aroclor 1016 or 1242.) Increased (P<0.05) mortality caused by P. berghei at 3 weeks. (No difference between Aroclor 1016 or 1242.) No histopathological changes in lung, thymus, mesenteric lymph nodes, spleen. Histopathological exam of line revealed hepatocytic hyperplasia.	
C57Bl/6	M/NR	18-20 g	Aroclor1016	Diet	167 mg/kg diet for 3 weeks	Splenic cells from treated mice injected into neonates elicited greater graft vs. host response, indicating Aroclor 1016 may activate donor lymphocytes.	Silkworth and Loose, 1978 (abstract)
Outbred albino	F/7/group	4-6 weeks	Aroclor 1248	Diet	0, 50, 100, 500, 1000 mg/kg diet for 3-5 weeks	PCB residues (mg/kg adiposa) 1.1, 109, 399, 1330, 3760 respectively at 3 weeks. No decreased food intake or other signs of toxicity except usual hepatocellular alterations.	Thomas and Hinsdill, 1978
	F/100/group				0, 1000 mg/kg diet for 5 weeks	At 4+ days postinoculation with live Salmonella typhimurium, treated mice had greater numbers of live organisms in liver and blood.	
	F/100/group				0, 100 or 1000 mg/kg diet for 5 weeks	Apparently dose-related increase in mortality due to S. typhimurium endotoxin.	
CL/ICR	F/ca 25/ group	Sexually mature	Kanechlor 500	Diet	0 or 500 mg/kg diet about 42 days	Depressed feed intake, hepatomegaly	Tanimura et al., 1980

TABLE V-3 (cont.)

Strain	Sex/No.	Weight/Age	Source of PCBs	Vehicle	Dose/Duration	Animal Effects	Reference
NMRI	F/45	Sexually mature	Clophen A-60	Peanut oil	0 or 0.025 mg/mouse daily for 62 days	Increased ($p<0.0005$) length of estrus cycle	Orberg and Kihlstrom, 1973
CD1	M/6/group	19-24 g	Aroclor 1254	Emulphor: Saline(1:8)	0, 10, 30, 100, 250 or 500 mg/kg bw, single dose	500 mg/kg: depression of spontaneous locomotor activity for <8 hours	Rosin and Martin, 1981

NR = Not reported

TABLE V-4

Acute and Short-Term Toxicity of PCBs Administered by Routes Other Than Oral

Route	Species/ Strain	Sex/No.	Weight/Age	Source of PCBs	Vehicle	Dose/Duration	Animal Effects	References
s.c.	Guinea pig/ NR	NR/54	NR	NR; 42% Cl	None	69-690 mg, single dose	Local injection necrosis to fibrous encapsulation. Hepatic: centrilobular necrosis, atrophy; fatty infiltration, splenic lymphoid hyperplasia, pulmonary congestion, mortality.	Miller, 1944
s.c.	Guinea pig/ NR	NR/10	NR	NR; ~42% Cl	Mineral oil	345 mg, single dose	As above; complete mortality 13 days; pulmonary congestion more severe	
s.c.	Rat/NR	NR/30	NR	NR; ~42% Cl	None	69 or 690 mg, single dose	Fatty liver degeneration; splenic hypertrophy; local injection fibrous encapsulation	
s.c.	Rabbit/NR	NR/3	NR	NR; ~42% Cl	None	690 or 1380 mg, single dose	Death in 14-72 days (as above, except liver contained fine droplets of fat).	
		NR/3	NR	NR; ~42% Cl	Mineral	345 or 690 mg, single dose	Death in 42-360 days (as above, except liver contained fine droplets of fat).	
dermal	Guinea pig/ NR	NR/11	NR	NR; ~42% Cl	None	34.5 mg/day for 11 days	Death within 21 days. (Dermal epithelial destruction. Lesions in internal organs as above.)	
		NR/16	NR	NR; ~42% Cl	Mineral oil	3.5-17 mg/day for 7 or 15 days		
s.c.	Mice/ddY	F/NR	7-8 weeks	Kanechlor 500	95% ethanol	0-10 mg/day for 10 days	Killed 2 days after last dose: Mortality ≥4 mg/day	Watanabe and Sugahara, 1981
i.p.	Rat/NR	M/NR	NR	Aroclor 1254	NR	100 mg/kg/ day for 6 days	Aminolevulinic acid (ALA) synthetase activity increased, ALA dehydratase activity decreased, microsomal heme and cytochrome P-450 increased	
inhalation	Rat/NR	M/5, F/5	NR	Decachlorobiphenyl	NA	2.54 g/m³ for 6 hours	Blinking and sneezing, reversible; 14-day observation, no effects on appetite or growth. Gross pathology, no lesions.	Berczy, 1974a

TABLE V-4 (cont.)

Route	Species/ Strain	Sex/No.	Weight/Age	Source of PCBs	Vehicle	Dose/Duration	Animal Effects	References
inhalation	Rat/NR	NR/4		Solvol	NA	10 g/m^3 for 3 hours	All uncoordinated, comatose, dead in 24 hours. Hepatic necrosis, fatty infiltration, renal tubules cloudy swelling. Heart and spleen congested, splenic necrosis.	Rozanora, 1943
inhalation	Rat/NR	NR/3		Solvol	NA	0.5 g/m^3 for 11 exposures	1 death, necropsy similar to above, signs less marked. Hyperplasia of Kupfer cells.	
i.p.	Rat/Wistar Woodlyn	M/7	young	pure mono-, di-, tetra-, hexa-, octa-chlorobiphenyls; pure mono-, di-, tetra-isomers of PCBs		50 mg/kg bw daily for 3 days	Hepatocytic proliferation of smooth endoplasmic reticulum, changes in rough ER, focal necrosis, cytoplasmic vacuolization.	Hansell and Ecobichon, 1974
						100 mg/kg bw daily for 7 days	More pronounced hepatic changes; fatty infiltration, centrilobular necrosis, biliary proliferation.	
i.p.	Rat/Wistar	M/NR	young	Aroclor 1016, 1221, 1232, 1242, 1248, 1254, 1260	Peanut oil	50 mg/kg/ day for 3 days	Induction of many hepatic enzymes.	Ecobichon, 1975
i.p.	Rat/Long-Evans	M/NR	3 weeks	Aroclor 1254 or 27 purified isomers	Corn oil	500 μmol/kg bw 1 time	Increased liver weight, decreased thymic weight; induction of various cytochrome P-450s	Parkinson, et al., 1983
i.p.	Rat/Sprague-Dawley	M/24	adult	Aroclor 1254	Corn oil	Control, corn oil, or 1 g/kg bw	"Typical" liver lesions; no effect on plasma corticosterone	Dunn et al., 1983
i.p.	Rat/Sprague-Dawley	M/NR	150-200 g	Aroclor 1254	Ringer's solution	0, 25, 50 mg/kg	Hepatomegaly, elevated cytochrome P-450, various hepatic enzyme systems induced or inhibited.	Hinton et al., 1978
i.p.	Rat/Sprague-Dawley	M/NR	100-120 g	Aroclor 1254	NR	100 mg/kg/day for 6 days	Elevated ALA synthetase, microsomal heme, cytochrome P-450. Depressed ALA dehydration.	Aharez and Kazzas, 1979
						100 mg/kg once weekly for 6 weeks	Elevated total porphyrins, microsomal heme, cytochrome P-450. Depressed ALA dehydration, ferrochelatase.	

TABLE V-4 (cont.)

Route	Species/ Strain	Sex/No.	Weight/Age	Source of PCBs	Vehicle	Dose/Duration	Animal Effects	References
i.p.	Rat/Sprague-Dawley	M/13	NR	Aroclor 1242	Peanut oil	100 mg/kg bw twice weekly for 6 weeks, once weekly for an additional 4 weeks	Body weight loss, hepatic midzonal sudanophilic vacuolization, focal necrosis, dilation of renal tubules with proteinaceous casts. Decreased PCV, RBC, hemoglobin, neutrophia. Increased serum iron, decreased corticosteroids and blood glucose. Increased urinary protein sugar, coproporphyrin.	Bruckner et al., 1974b
i.p.	Rat/Sprague Dawley	M/24	200-300 g	Aroclor 1242	Peanut oil	0, 1, 5, 25, 50 or 100 mg/kg bw,	Elevated hydroxylation, N-demethylation, cytochrome P-450	Bruckner et al., 1974b
		M/6/treated group, 3/ control group	NR	Aroclor 1242	Peanut oil	single dose 0 or 100 mg/kg bw, single dose	Examinations of microsomal enzyme induction at 1, 5, 10, 20, 40 days post-treatment indicated maximum induction at 5 days, some residual induction at 20 days. Hydroxylation most dramatically elevated.	
i.p.	Rat/Wistar	M/120	250 g	Clophen A-50	Corn oil	0 or 100 mg/kg bw, single dose 4 weeks observation	Cytochrome P-450 increased 3- to 4-fold, maximum in 1 week. NADPH activity doubled, P-nitroanisole O-demethylase induced 6- to 7-fold; ~4-fold after 1 month. AHH activity increased 3-fold, down to normal ~1 month. Microsomal epoxide hydratase increased 2.5-fold at 1 week, persisted at least 4 weeks. Glutathione S-transferase increased at 1 day remained at these levels. Microsomal UDP glucuronosyltransferase activity increased 2.5-fold in 1 week, persisted 4 weeks.	Parkki et al., 1971
i.p.	Mice/BALB/CJ	NR/4-10/ group	NR	Aroclor 1242	Corn oil	1000 mg/kg bw single dose	Splenomegaly: significant by day 6, peak by day 9, gone by day 13. Cellularity: significant reduction in lymphocytes days 6-10.	Carter and Clancey, 1980

TABLE V-4 (cont.)

Route	Species/ Strain	Sex/No.	Weight/Age	Source of PCBs	Vehicle	Dose/Duration	Animal Effects	References
dermal	Rabbit/New Zealand White	F/16	2500-3050 g/5 months	Phenoclor DP6 Clophen A-60 Aroclor 1260	Isopropanol	118 mg/day, 5 days/week for 38 days (27 applications)	All PCBs: Gradual weight loss, mortality beginning at day 10, erythemia and thickening of skin, subcutaneous edema, ascites, UV fluorescence. Fecal coproporphyrin and protoporphyrin elevated. Hematology: leukopenia. Apparent hepatomegaly, kidney enlargement. Liver: centrilobular degeneration, focal hydropic degeneration, focal necrosis, centrilobular hepatocytic atrophy (more pronounced in Clophen, least in Aroclor group), periportal fibrosis. Kidney: hydropic degeneration of convoluted tubules, punotic nuclei, rhexis and lysis of tubular epithelial, cells, tubular dilatation with costs of necrotic cells.	Vos and Beems, 1971
dermal	Rabbit/New Zealand White	F/12	2.5-2.9 kg/3.5 months	2,4,5,2',4',5'-HCB Aroclor 1260	Isopropanol	120 mg/day, 5 days/week for 4 weeks (20 applications)	Dermal: erythema, wrinkling, hyperkeratosis, reduced hair regrowth; more severe in Aroclor 1260 group. Fecal coproporphyrin elevated in both treatment groups with UV fluorescence. Hepatomegaly, elevated SGPT, SGOT in 2,4,5-HCB group. Dermal sections: epidermal follicular hyperplasia; follicular plugging more in Aroclor 1260 group. Moderate thymic atrophy in both PCB groups. Liver lesions similar to previous study (Vos and Beems, 1971), more severe in 2,4,5-HCB group; SER proliferation, RER dilation and degranulations, picnotic nuclei.	Vos and Notenboom-Ram, 1972

NR = Not reported

TABLE V-5

Acute and Short-Term Toxicity of Orally-Administered PCBs to Other Species

Species/ Strain	Sex/No.	Weight/Age	Source of PCBs	Vehicle	Dose/Duration	Animal Effects	Reference
Monkey/ rhesus	M/7	9.2 kg/ adult	2,5,2',5'-tetra-CB	corn oil	0.18 mg/kg bw	Moderate proliferation of SER, slight decrease of liver DNA, increase in cytochrome P-450.	Allen et al., 1975
Rabbit/NR	F/16	adult	Aroclor 1254	corn oil	0, 1.0, 10, 12.5, 25, 50 mg/kg bw daily for 28 days	No effect on total number of fetuses, number of viable fetuses, number of resorption sites or number of abortions at doses of 1.0 or 10 mg/kg bw/day. Liver weights in the dams were significantly increased at the 10 mg/kg bw/day dose. Effects on fetal viability as well as other maternal effects were seen at doses of 12.5 or greater.	Villeneuve et al., 1971
Rabbit/New Zealand White	M/7/group	~2 kg	Aroclor 1254	diet	0, 3.7, 20.0, 45.8, 170 mg/kg diet for 8 weeks: 0.18, 0.92, 2.1 or 6.54 mg/kg bw/day, respectively	No effect on feed consumption, growth rate, visceral pathology except apparent hepatomegaly at all levels which was statistically significant (p=0.05) at the highest two doses. No effect on hematologic parameters. No consistent immunological response (hemolysis or hemagglutination titers) to sheep RBC. Gamma globulin reduced at all levels. No effect on dermal tuberculin reaction. Splenic and thymic reductions in gamma globulin-producing cells, dose-related.	Street and Sharma, 1975
Guinea pig/ Dunbar/ Hartley	F/5-19/ group	500-600 g/ 8-10 weeks	Clophen A-50 2,2',4,4',5,5'-HCB	peanut oil	25 or 100 mg. Total doses over 55 days	"All seemed unaffected by the treatment." Clophen A-50: increased cytochrome P-450	Brunstroem et al., 1981
Guineapig/ NR	M/40	3-4 weeks/ ~225 g	Clophen A-60	diet	0, 10, 50, 250 mg/kg diet for 4 weeks	250 mg/kg: 80% mortality; cachexia, lymphoid atrophy, liver damage, hepatomegaly ≥50 mg. ≥10 mg/kg: dose-related reduction in hemagglutination titers and tetanus antitoxin-producing cells in popliteal lymph nodes following tetanus toxoid immunization.	Vos and van Genderen, 1973
	F/40	3-4 weeks/ ~225 g	Clophen A-60	diet	0, 10, 50 mg/kg diet for 6 weeks	50 mg/kg Clophen: increased liver weight.	
			Aroclor 1260	diet	50 mg/kg diet for for 6 weeks	Significant reduction of tetanus antitoxin titers, circulating leukocytes and lymphocytes and thymus atrophy with both PCBs, 50 mg/kg diet. 100% mortality at 250 mg/kg. Reduced tuberculin skin reaction, thymus atrophy, leukopenia at 50 mg/kg level	
	F/30		ClophenA-60	diet	0, 50, 250 mg/kg diet for 7 weeks.		

TABLE V-5 (cont.)

Species/ Strain	Sex/No.	Weight/Age	Source of PCBs	Vehicle	Dose/Duration	Animal Effects	Reference
Guinea pig/ albino	F/36	~220 g/ 4 weeks	Aroclor 1260	diet	0, 10, 50 mg/kg diet for 8 weeks	50 mg/kg: reduced rate gain, hepatomegaly, reduced weight of kidney, adrenals. 10 mg/kg: splenic atrophy; reduced popliteal lymph node, gammaglobulin-producing cells.	Vos and de Roij, 1972
Pig/ Yorkshire	NR/11	14 days	Aroclor 1254	condensed milk added to diet	0, 12.5, 25, 50, 100 mg/kg bw up to 35 days	All levels: partial anorexia, reduced weight gain, diarrhea (melena or blood at ≥50 mg/kg), abdominal distension, gastritis, colitis, enlarged thyroid, thymic and splenic atrophy, liver and kidney enlargement.	Miniats et al., 1978
Monkey/ rhesus	F/12	5.6 kg/7-10 years	Aroclor 1248	diet	0, 25 mg/kg diet for 2 months; total intake of 260-450 mg	Alopecia; edema of lips, eyelids, face; pustules involving hair follicles; pruitis; necropsy of the animal consuming 450 mg: severe weight loss, subcutaneous edema, acute hyperplastic gastritis with focal hemorrhage, ulceration. Liver: focal necrosis, enlarged hepatocytes, lipid accumulation. Hypocellularity of bone marrow.	Allen et al., 1974
Monkey/ rhesus	M/7	9.2 kg/adult	2,5,2',5'-TCB	corn oil	0 or 18 mg/kg bw, single dose	"No obvious clinical effects." No change in hematology. Levels (14 days) of TCB: adiposa, 135; adrenals, 33; lung and heart, 9; liver and skin, 4; thyroid, 2 μg/g tissue. Microscopically, all tissues normal. Hepatic ultrastructive: proliferation of SER, elevated cytochrome P-450.	Allen et al., 1975
Monkey/ rhesus	NR	adult	Aroclor 1248	diet	100 or 300 mg/kg diet for 2-3 months	Gradual weight loss, alopecia lacrimations, conjunctival congestion, facial edema, comedones, large intrafollicular keratin cysts. Hematology: gradual decrease in PCV, Hb; lymphocytopenia and concomitant neutrophilia. Reduced serum proteins, lipids, cholesterol, triglycerides. Thickened gastric mucosa with mucin-filled cysts, moderately invasive gastric hyperplasia. Two-fold hepatomegaly, enlarged hepatocytes with increased smooth endoplasmic reticulum; decreased liver DNA, RNA, increased MFO.	Allen, 1975

TABLE V-5 (cont.)

Species/ Strain	Sex/No.	Weight/Age	Source of PCBs	Vehicle	Dose/Duration	Animal Effects	Reference
Monkey/ rhesus	M/2 F/2	2.8-3.6 kg	2,5,4'-TCB	diet	5 mg/kg diet for 84 days (historic controls)	Most organs: increased blood volume. Increased relative liver, brain weight (males). Microscopically, venous congestion in many tissues. Adrenals: hemorrhages and cellular changes in zona fasciculata. Renal cortical and medulary degeneration, tubal colloid. Liver: congestion, fatty infiltrations. CNS: parenchymal and mesenchymal degeneration, macrozlial proliferation, gliosis, swelling of Purkinje cells.	Iatropoulos et al., 1977

NR = Not reported

Acute and Short-Term Toxicity in the Rat -- Many studies of PCB-induced toxicity in the rat were performed using the oral route. The studies discussed in the following sections are also summarized in Tables V-2, etc. Additional details can be found in these tables.

Gavage treatment with 0.05 g Aroclor 1254/kg bw to groups of four male Sprague-Dawley rats weighing 200-250 g daily for 21 days resulted in depressed food and water intake, depressed body temperature and reduced rate of gain or frank weight loss (Komives, 1979; Komives and Alayoku, 1980). Similar results were obtained by Kling et al. (1978) and Garthoff et al. (1977) using similar doses of Aroclor 1254 in the diet. Other workers (Chu et al., 1977), however, reported that exposure of male Sprague-Dawley weanling rats to 20 mg Aroclor 1254 or Aroclor 1260/kg diet for 28 days did not reduce feed intake or rate of weight gain. Oishi et al. (1978) likewise reported that 100 mg/kg Kanechlor 500 in the diet failed to reduce feed or water intake in rats.

The majority of the acute toxicity studies have evaluated the effects of PCBs on the liver. The details of these effects are discussed in the Target Organ Toxicity Section. Grant and Phillips (1974) administered Aroclor 1254 by gavage to male (40) and female (44) Wistar rats. In yearling males levels as low as 12.5 mg/kg bw/day for 7 days significantly increased liver weights. In females 30-120 days old, 5 mg/kg bw/day for 7 days increased liver weights. More recently, Baumann et al. (1983) reported that groups of 10 male Wistar/Neuherberg rats treated with Clophen A-50 by gavage twice weekly for 6 weeks exhibited liver damage at levels as low as 2 mg/kg bw/day. Liver damage was manifested as increased liver size, congestion, fatty degeneration and focal necrosis. Additionally, changes in blood chemistry were noted. Lin et al. (1979) did not report increased liver size, but found considerable ultrastructural damage (intracellular vacuolization, fragmentation of rough endoplasmic reticulum, altered mitochondrial conformation) in three 200-250 g female Sprague-Dawley rats that were treated by gavage with single doses of 1.7 g 2,5,2',5'-TCB/kg bw.

PCBs have been used widely to induce hepatic enzymes, often in studies with other chemicals. In these studies, large doses of PCBs are often given by i.p. injection or gavage to obtain maximal enzyme induction. Fewer studies, however, have been designed to define minimum effective oral doses. Chu et al. (1977) reported induction of MFO activity in male weanling rats exposed to 20 mg Aroclor 1254 or Aroclor 1260/kg diet for 28 days. Similarly, Garthoff et al. (1977) reported that 5-50 mg Aroclor 1254/kg diet for 2 weeks resulted in induction of many liver enzyme systems in groups of 6-16, 250 g male Holtzman rats.

PCBs have been shown to cause splenic, thymic and lymph node regression (Allen et al., 1975; Allen and Abrahamson, 1973, Parkinson et al., 1983), although some workers (Miller, 1944; Oishi et al., 1978) reported slight splenic enlargement. Aroclor 1254 at levels as low as 5 mg/kg diet for 4 weeks has been shown to cause considerable change in the microscopic and ultrastructural appearance of the thyroid gland in groups of six male 8-week-old Osborne-Mendel rats (Collins and Capen, 1980b). The investigators noted that Aroclor interfered with thyroid function and reduced thyroxine.

Other effects of PCBs on endocrine function have been reported. Dietary administration of 300 mg Aroclor 1254/kg diet for 10 days to 56, 200-300 g female Holtzman rats (Spencer, 1982) has been associated with decreased levels of placental protein and glycogen content. Gavage treatment of 18 male Sprague-Dawley rats (250-300 g) with 50 mg Arochlor 1254/kg bw for 7 days resulted in increased acid phosphatase activity in testicular interstitial cells (Dikshith et al., 1975). These studies may indicate that PCBs indirectly hasten steroid catabolism (Spencer, 1982).

In general, blood and urine chemistries appeared to be affected by oral administration of various PCB products (Oishi et al., 1978). Blood levels of glucose were reduced and blood levels of urea nitrogen, cholesterol and protein were increased by 50 mg Aroclor 1254/kg diet fed for 2 weeks to groups of 6-16 male 250 g Holtzman rats (Garthoff et al., 1977). Baumann et al. (1983) demonstrated increased levels of urinary porphyrin and porphyrin precursors in groups of 10, 100 g male Wistar/Neuherberg rats following treatment by gavage with 50 mg Clophen A-50/kg bw for 6 weeks.

Only one study was found that implicated PCBs (Aroclor 1254) in dermatitis in rats (Zinkl, 1977). Alopecia and a crusty dermatitis with serum ooze developed first on the ears, then the dorsum of the nose, tail and feet of female CD rats after 10 weeks of exposure to 100 mg Aroclor 1254/kg diet.

Acute and Short-Term Toxicity in the Mouse -- Toxic effects of PCBs in mice parallel those observed in rats. These studies are summarized in Tables V-3 and V-4. Depressed food intake and rate of weight gain and lethargy were reported (Tanimura et al., 1980; Sanders et al., 1974). A dose as low as 8 mg of 3,4,5, 3',4',5'-HCB/kg bw/day given by gavage for 10 days to pregnant CD-1 mice caused the effects described above (Marks et al., 1981). Dietary exposure of five adult male ICR mice to 250 mg Aroclor 1254/kg diet for 14 days also resulted in depressed food intake (Sanders et al., 1974). Pure 3,4,5,3',4',5'-HCB appeared to be more highly toxic than Aroclor 1254 on an equal weight basis. Carter and Cameron (1977), however, observed no effect on body weight or food or water intake in groups of 10 male Swiss albino mice that were exposed to 1000 mg of 2,4,5,2',4',5'-HCB/kg bw by gavage daily for 28 days.

Effects on the livers of mice are strikingly similar to those observed in rats (Carter and Cameron, 1977; Orberg, 1978; Sanders et al., 1974; Loose et al., 1978a,b). Levels of 0.3-1.0 mg of 3,4,5,3',4',5'-HCB/kg diet for 28 days resulted in liver enlargement; fatty degeneration and formation of microabscesses (Biocca et al., 1981) were found at doses of 1.0 mg/kg of diet or greater. This study used groups of 5 C57Bl/6J male mice to test the relative potency of four symmetrical isomers of HCB. The other isomers tested elicited similar responses at the 30-300 mg/kg diet level. Mattsson et al. (1981) observed hepatomegaly in groups of 10 female CBA mice that were exposed to 2,4,5,2',4',5'-HCB in peanut oil at levels of 0.5 mg/mouse/day for 7 days. Higher levels of commercially available PCB products are necessary to elicit these hepatic responses. Loose et al. (1978b) exposed groups of ~13 male BALB/CJ mice to 167 mg Aroclor 1242/kg diet for 6 weeks to demonstrate hepatocytic hypertrophy. Sanders et al. (1974) demonstrated hepatomegaly in groups of five adult male ICR mice exposed to dietary levels of 250 mg/kg Aroclor 1254 for 14 days.

Mice have been used as a model to demonstrate the effects of PCBs on suppression of the immune system. Using increased mortality to *Salmonella typhosa* endotoxin and *Plasmodium berghei*, Loose et al. (1978a,b) demonstrated immunosuppression in groups of ~13 male BALB/CJ mice that were treated with 167 [illegible] Aroclor 1016 or 1242/kg diet for 6 weeks. These treatments did not result in histologically-demonstrable lesions in thymus, spleen or mesenteric lymph nodes. Thomas and Hinsdill (1978) demonstrated decreased mortality of *Salmonella typhimurium* in groups of seven outbred female mice that were given 1000 mg Aroclor 1248/kg diet for 5 weeks and an apparent dose-related increase in mortality caused by *S. typhimurium* endotoxin at levels of 100 and 1000 mg/kg diet.

Acute and Short-Term Toxicity in Other Species -- Manifestations of PCB toxicity in the rabbit resembled those in other species (Villeneuve et al., 1971) with the exception that dermal application of any of three commercially available PCB products resulted in severe renal damage (Vos and Beems, 1971). Applied at 118 mg, 5 times weekly for 27 applications (38 days), Phenoclor DP6, Clophen A60 or Aroclor 1260 all resulted in hydropic degeneration of convoluted tubules, destruction of tubular epithelial cells with resultant tubular dilatation and proteinaceous casts. No mention of such lesions was made in a subsequent study by this laboratory using a total of 20 such applications (Vos and Notenboom-Ram, 1972). Oral administration of levels as low as 3 mg Aroclor 1254/kg diet for 8 weeks to seven male New Zealand rabbits failed to produce signs of toxicity other than hepatomegaly and immunosuppression as evaluated by splenic and thymic reduction of gamma globulin-producing cells (Street and Sharma, 1975).

Guinea pigs may be the most sensitive laboratory species to PCB toxicity. Vos and van Genderen (1973) reported 80 and 100% mortality of groups of four male and female guinea pigs that were exposed to 250 mg Clophen A60/kg diet for 4-7 weeks. Weight loss, atrophy of lymphoid tissue and hepatomegaly with "liver damage" were reported. However, recently Brunstroem et al. (1981) reported that all animals "seemed unaffected by the treatment" in a study that exposed 5-19 pregnant females to total Clophen A50 or 2,4,5,2',4',5'-HCB amounts of 100 mg over a 55-day period. In this species, one laboratory (Vos and de Roij, 1972; Vos and van Genderen, 1973) demonstrated immunosuppression resulting from a 4- or 6- to 7-week exposure of groups of 30-40 female albino guinea pigs to 50 mg of Clophen A60 or Aroclor 1260/kg diet.

Signs of toxicity in monkeys acutely exposed to PCBs closely parallel those reported in other species with a few notable exceptions. Frequently, the first obvious sign of PCB intoxication is the development of facial lesions. Facial and palpebral edema followed by alopecia and development of comedones was reported in six female rhesus monkeys exposed to 25 mg Aroclor 1248/kg diet for 2 months (Allen et al., 1974b; Allen, 1975). Total Aroclor 1248 intake was calculated on the basis of known food intake to be ~260 mg. These signs progressed to generalized subcutaneous edema, pruitis and alopecia. As described in more detail in Table V-8, chronic low level toxicity studies by McNulty et al. (1980) resulted in lesions identical to those reported for rhesus monkeys fed higher doses of Aroclor 124F.

These workers also reported the development of gastritis progressing to a moderately invasive gastric hyperplasia in the individuals described in the study cited above (Allen et al., 1974b) and in six male monkeys exposed for 3 months to a diet containing 300 mg Aroclor 1248/kg diet (Allen and Norback, 1973). Within 1 month, all the PCB-exposed animals suffered from alopecia, subcutaneous edema, purulent ocular discharge and acneform lesions. Upon necropsy, edematous thickening of the stomach wall accompanied by glandular hyperplasia was observed. Glandular cells accumulated mucus, resulting in the formation of large, mucus-containing cysts. Alterations of the glandular epithelial cells and their nuclei accompanied by inflammatory processes and invasion of the muscularis were observed.

Iatropoulos et al. (1977) exposed 2 male and 2 female rhesus monkeys to 5 mg of 2,5,4'-TCB/kg diet for 84 days. No mention of total PCB intake was made. They reported a generalized increased blood volume of many tissues apparently resulting from dilation of arterioles, capillaries and veins. Hemorrhages and cellular changes in the adrenal cortex were observed. They also reported parenchymal and mesenchymal degeneration in the brain.

Chronic and Subchronic Toxicity. Chronic and subchronic toxicity studies discussed in this section include those >90 days in duration. These studies are summarized in Tables V-6 to V-9. Since more chronic toxicity studies have been performed using rats, they will be the first species discussed. Chronic toxicity in other species will then be compared with toxicity in the rat.

TABLE V-6

Effects of Subchronic and Chronic Oral Exposure of PCBs to Rats

Strain	Sex/No.	Source of PCB	Vehicle	Dosage Schedule	Duration of Study	Animal Effects	Reference
F344	M,F/191	Aroclor 1254	diet	0, 25, 50, 100 mg/kg diet for 2 years	2 years	Reduced body weight. Stomach: "intestinal" metaplasia, dose-related; adenocarcinoma of glandular stomach.	Morgan et al., 1981
Sherman	F/400	Aroclor 1260	diet	0 or 100 mg/kg diet for 21 months	21 months	No effect on food intake; reduced body weight. Elevated tan liver modules (170/184), hepatocellular carcinoma (26/184). Other areas: hepatic disruption.	Kimbrough et al., 1975
Wistar	M/290	Kanechlor 300, Kanechlor 400 or Kanechlor 500	diet	0, 500 or 1000 mg/kg diet for 27-52 weeks	1 year	Heavy mortality. Hepatomegaly, oval cell and bile duct proliferation, fatty liver infiltration. Cholangiofibrosis at 1000 mg/kg level of all three Kanechlors, nodular hyperplasia. Depressed final body weight.	Ito et al., 1974
Sprague-Dawley	M/6/group	Aroclor 1242	diet	0, 5 or 25 mg/kg diet for 2, 4 or 6 months	2, 4 or 6 months	Elevated hepatic microsomal enzyme activity, lipid content. Elevated urinary coproporphyrin levels. Present after 2 months at 5 mg/kg.	Bruckner et al., 1974a
Sprague-Dawley	M/96	Aroclor 1248, Aroclor 1254 or Aroclor 1262	diet	0 or 100 mg/kg diet for 52 weeks	65 weeks	Increased hepatic protein, RNA and lipid; decreased DNA. Increased microsomal total protein and cytochrome P-450. Induced N-demethylase nitroreductase. Inhibited glucose-6-phosphatase.	Allen and Abrahamson, 1973
CD	F/300	Aroclor 1254	diet	0, 10, 30 or 100 mg/kg diet for up to 20 weeks	20 weeks	Serum cholesterol, beta globulin increased, gamma globlin decreased (dose-related). ≥30 mg/kg: Reduced rate of gain, hepatomegaly, cardiomegaly (dose-related). ≥10 mg/kg: Hepatic porphyrinic fluorescence. ≥10 mg/kg: Erythema, crustiness,2 hyperkeratosis, perikeritosis on ears, dorsum of nose and feet, tail.	Zinkl, 1977
Donryu	M/15, F/15	Kanechlor 400	diet	total intake 450-1500 mg over 159-560 days	560 days	All treated rats: fatty liver degenration. Females 1200-1500 mg: multiple adenomatous nodules. All rats >700 mg: hepatomegaly. Lung and intracranoid abscesses suggested impaired resistance to infection.	Kimura and Baba, 1973

TABLE V-6 (cont.)

Strain	Sex/No.	Source of PCB	Vehicle	Dosage Schedule	Duration of Study	Animal Effects	Reference
Sprague-Dawley	F/6/group	Aroclor 1242	diet	0, 75 or 150 mg/kg diet for 8 or 36 weeks	36 weeks	Both levels: massive venous engorgement of liver with characteristic darkening; marked focal neurosis and regeneration, enlarged hepatocytes; many mitoses and multinucleate cells, accumulation of pigment adjacent to veins, heaviest in Kupfer cells; accumulation of lipid droplets in cytoplasm, some with areas suggestive of lipid-cholesterol complexes; marked SER proliferation; deposits of iron; granular degeneration of mitochondria; many hepatocytes contained whorl-like membranous bodies.	Jonssen et al., 1981
Sherman	M/10, F/10 per group	Aroclor 1254	diet	0, 20, 100, 500 mg/kg diet for 8 months	8 months	Mortality (3/20) and reduced rate of gain at 500 mg/kg. Hepatomegaly, enlarged hepatocytes with foamy cytoplasm-containing inclusions at ≥20 mg/kg. Adenofibrosis and pigment accumulation at ≥100 mg/kg.	Kimbrough et al., 1972
	M/10, F/10 per group	Aroclor 1260	diet	0, 20, 100, 500, 1000 mg/kg diet for 8 months	8 months	Mortality 1/10, 2/10, 8/10 of females in 100, 500, 1000 mg/kg groups. Decreased rate of gain at ≥500 mg/kg. Hepatomegaly, male and female at ≥20 mg/kg, discolored livers with UV fluorescence, enlarged hepatocytes with foamy cytoplasm-containing inclusions. Increased lipid content at ≥100 mg/kg. Pigment accumulations at 500 mg/kg. Adenofibrosis at ≥100 mg/kg.	
Sprague-Dawley	M/96	Aroclor 1248, Aroclor 1254 or Aroclor 1262	diet	0 or 100 mg/kg diet for 52 weeks	65 weeks	Normal appetites, appearance, weight gain, Hb, PCV, WBC, serum protein, A/G ratios. Elevated serum total lipids, cholesterol. Total lipid and triglycerid spiked very high peaks on Aroclor 1254 (only) at 52 weeks. Cholesterol levels persisted at 65 weeks (13 weeks off exposure). Triglyceride levels fell less than controls by 65 weeks. Hepatomegaly; focal degeneration and necrosis by 13 weeks.	Allen et al., 1976

TABLE V-7

Subchronic and Chronic Oral Exposure of PCBs to Mice

Strain	Sex/No.	Source of PCBs	Vehicle	Dosage Schedule	Duration of Study	Animal Effects	References
Mice/dd	M/114	Kanechlor 300, Kanechlor 400 or Kanechlor 500	diet	0, 100, 250 or 500 mg/kg diet for 32 weeks	32 weeks	Hepatomegaly in all treatment groups. No effect on final body weight. Oval cell formation and bile duct prolifera- at mid- and high-dose levels of Kanechlor 400 and Kanechlor 500. Hepatocellular hypertrophy in all exposed groups except low-dose Kanechlor 300. Amyloidosis in all exposed groups except high-dose Kanechlor 400 and Kanechlor 500 groups; greater incidence associated with lower doses of doses of lower-chlorinated Kanechlors.	Ito et al., 1973
BALB/CJ	M/25/group	Aroclor 1221, Aroclor 1242 or Aroclor 1254	diet	0, 3.75, 37.5 or 375 mg/kg diet for 6 months	9 months	375 mg/kg Aroclor 1242 and all Arochlor 1254-exposed groups: significant ($p<0.01$) increase in liver weight after 6 months. Significant ($p<0.01$) decrease in liver weight of 375 mg/kg Aroclor 1242 and 37.5 mg/kg Aroclor 1254-exposed groups after 3-month recovery period. Arochlor 1254: 375 mg/kg: mortality and severe hepatopathology; 37.5 mg/kg: mild hepatopathology; 3.75 mg/kg: no liver lesions. Aroclor 1242: 375 mg/kg: moderate hepatopathology. Aroclor 1221: no liver lesions.	Koller, 1977
Swiss Albino	F/63	Aroclor 1254	diet	200 mg/kg diet for 23 weeks	23 weeks	Thickening and erythemia of pinara of the ear; changes in microvascular.	Bell, 1983
BALB/CJ	M/200	Aroclor 1254	diet	0 or 300 mg/kg diet for 6 or 11 months	11 months	Both 11- and 6-month exposure: Hepatomegaly, hepatomas, liver degeneration and elevated porphyrin.	Kimbrough and Linder, 1974
ddN	F/60	"CBP"	olive or rice bran oil	0, 0.5% V/V CBP in olive oil or 1600 mg/kg CBP in bran oil for 13, 17, 22 or 26 weeks	26 weeks	Both exposed groups: slight weight loss, reduced activity; eczematous and ulcerative skin lesions, hepatomegaly and hepatopathology.	Nishizumi, 1970

TABLE V-8

Effects of Chronic and Subchronic Exposure of PCBs to Monkeys

Strain	Sex/No.	Source of PCBs	Vehicle	Dosage	Duration of Study	Animal Effects	Reference
Rhesus	F/24 M/NR	Aroclor 1248	diet	0, 2.5 or 5.0 mg/kg diet -18 months	-39.6 months	Males (5.0 mg/kg level only): moderate erythema and periorbital edema. Females: more severe skin lesions (alopecia, acne); extreme weight loss, irregular menstrual cycle length, depressed serum progesterone. Considerable improvement after 1-year recovery period.	Barsotti and Allen, 1975
Rhesus	F/30	Aroclor 1248	diet	0, 2.5 or 5.0 mg/kg mg/kg diet	-16 months Total intake 90 or 180 mg/kg by females in 6 months	Skin lesions as above; 15% weight loss in females. Normal hematograms. After 6 months: serum total lipids reduced, shift in A/G ratio, elevated SGPT. Menstrual cycles lengthened. Serum progesterone and estradiol reduced. After 12 months, serum cholesterol and triglyceride reduced.	Barsotti, et al., 1976 Allen, et al., 1979a Barsotti, 1981
	M/10	Aroclor 1248	diet	0 or 5.0 mg/kg diet about 16 months			
	F/8/group	Aroclor 1248	diet	0.5 or 1.0 mg/kg diet 3 times weekly for -16.6 months	-16.6 months Total intake -8 or 16 mg after 7 months	No irregularities in menstrual cycle, or serum estradiol, progesterone or reproduction success. Infants smaller, skin hyperpigmentated.	
	F/24	Aroclor 1016	diet	0.025, 0.25 or 1.0 mg/kg diet for unreported length of time	NR	No abnormalities of clinical, gross or reproductive parameters. 1.0 mg/kg: Infants reduced birthweight.	
Rhesus	NR/7	Aroclor 1248	transplacental or mother's milk	Mothers exposed to PCBs 6 months prior to gestation, through gestation and 3-4 months of nursing	Up to infant age of 24 months	Significantly increased locomotor behavior (hyperactivity). Significantly retarded learning ability.	Bowman et al., 1978, 1981
		Aroclor 1248	transplacental or mother's milk	Mothers removed from exposure to PCBs for 22-84 weeks before conception		Mothers exposed to ≥ 2.5 mg/kg: hyperactivity.	

TABLE V-8 (cont.)

Strain	Sex/No.	Source of PCBs	Vehicle	Dosage	Duration of Study	Animal Effects	Reference
Rhesus	M/3/group	3,4,3',4'-TCB or 2,5,2',5'-TCB	diet	3 mg/kg reduced to 1 mg/kg reduced to to 0.3 mg/kg	Up to 215 days	Mortality of all three by day 215 in 3,4,3',4'-TCB-exposed groups: emaciation, skin lesions, nail bed hyperplasia, loss of nails, thymusatrophy, gastric lesions as described (Allen, 1975). 2,5,2',5'-TCD: no signs of toxicity, no gross or histologic lesions.	McNulty et al., 1980
		Control		Elevated to 1 mg/kg			
	M/4-5/group	3,4,3',4'-TCB or 2,5,2',5'-TCB	diet	1 mg/kg for 38 days or 133 days, then control diet	~190 days	1 death: necropsy findings as above. Others: squamous metaplasm of sebaceous glands.	
		Aroclor 1242		1 mg/kg for 133 days 5 mg/kg additional 2 month		No evidence of toxicity.	
Rhesus	M/6	Aroclor 1242	diet	0, 3, 10, 10, 30 or 100 mg/kg diet	Up to 245 days	All PCB-exposed monkeys: palpebral swelling, erythemia; weight loss, rough hair coat, reduced Hb, leukocytosis. Mortality of 4/6 by day 245. Gastric lesions: hypertrophic gastric mucosa consisting of elongated hyperplastic glands, destruction of parietal and zymogenic cells. Only specific region along greater curvature affected.	Becker et al., 1979
Cynomolgus	F/4	Aroclor 1254	corn oil, gelatin, apple juice	0, 100, 100 or 400 mg/kg bw/day, 3 days/week	Up to 238 days	Lost fingernails, fetal toxicity. Substantially reduced antibody production to SRBC antigen.	Truelove et al.,1982
Cynomolgus	F/7	P-KC-400^a Y-PCB^b	olive oil in banana	5 mg/monkey/day 5 or 10 mg/monkey/day	20 weeks	Death of 10 mg Y-PCB-dosed monkeys by 8 weeks. Weight loss of P-KC-400 and 5 mg Y-PCB-dosed monkeys. Y-PCB-dosed: aloperia, acne, hyperpigmentation, periorbital edema. All treatments: reduced antibody production to SRBC. Histopathology Y-PCB: enlarged hepatocytes with enlarged SER, focal necrosis, bile duct proliferation. Dilated renal tubules with casts, epithelialvacuoles. Meibomian cysts, skin hyperkeratosis. Histopathology P-KC-400 and PY-PCB: lesions in liver, kidney as above, but more mild. Periorbital skin: no lesions.	Hori et al., 1982
		PY-PCB^c		5 mg/monkey/day			
		control		NA			
				All above treatments given 6 days/week			

TABLE V-8 (cont.)

Strain	Sex/No.	Source of PCBs	Vehicle	Dosage	Duration of Study	Animal Effects	Reference
Rhesus	M/6	"PCB"	salad oil in banana	0 or 0.5 mg/kg bw (79.2-253.6 mg)	Up to 5 months	Symptoms of all treated animals similar: 17.3% weight loss, partial alopecia, palpebral edema, acneform eruptions, squamous metoplasia of meibomian glands.	Ohnishi and Kahno, 1979
	F/6	"PCB with PCDF"	diet	2.5 mg/kg bw PCDF			

[a] P-KC-400 = Kanechlor 400 with PCDFs removed

[b] Y-PCB was prepared from Kanechlor 400, contained ~400 ppm PCDFs

[c] PY-PCB = Y-PCB with PCDFs removed

NA = Not available

TABLE V-9

Effects of Subchronic and Chronic Oral Exposure of PCBs to Other Species

Strain	Sex/No.	Source of PCBs	Vehicle	Dosage Schedule	Duration of Study	Animal Effects	References
Mink/ Pastel	M,F/105	Aroclor 1242	diet.	0, 5, 10, 20 or 40 mg/kg diet	8 months	Mortality of all mink on ≥20 mg/kg diet.	Bleavins et al., 1980
		Aroclor 1016	diet	0 or 20 mg/kg diet		Death of 3/12 (25%) of females. Necropsy: emaciation, complete absence of body fat, gastric ulceration. Aroclor 1242 at 5 or 10 mg/kg diet: complete reproductive failure. Aroclor 1016 at 20 mg/kg diet: reduced reproductive performances.	
Mink/NR	F/7 or 8/ group	Aroclor 1016, Aroclor 1221, Aroclor 1242 or Aroclor 1254	diet	0 or 2 mg/kg diet	10 months	Aroclor 1254: interference with reproduction. All Aroclors tested: no significant differences in body weight gain, hemoglobin, PCV.	Aulerich and Ringer, 1977
Rabbit/ New Zealand White	F/20, M/20	Aroclor 1221, Aroclor 1242 Aroclor 1254	corn oil	0 or 300 mg 1 time weekly for 14 weeks	14 weeks	Aroclor 1242, 1254: elevated SGOT, SGPT. Aroclor 1254: slight transient increase in serum cholesterol, reduced rate of gain, severe hepatomegaly, uterine atrophy. No differences in hematologic parameters, BUN, serum protein fractions. Histopathology, Aroclor 1254: vacuolated and granular enlarged hepatocytes, centrilobular necrosis and fibrosis. Ballooned RER. Lesions less obvious in Aroclor 1242-exposed, absent in Aroclor 1221-exposed.	Koller and Zinkl, 1973

NR = Not reported

Chronic and Subchronic Toxicity in the Rat -- Oral chronic and subchronic toxicity studies in the rat are summarized in Table V-6. Morgan et al. (1981) reported on the toxicity of feeding diets containing Aroclor 1254 to 191 male and female F344 rats for 2 years. Rats were exposed to levels of 0, 25, 50 or 100 mg of Aroclor 1254/kg of diet. Mortality occurred in 8 and 33; 17 and 21; 42 and 17; and 54 and 29% of the males and females, respectively, in these groups. Mean final body weight of all treatment groups were lower than the body weight of the control groups with the exception of the low-dose group males. Beginning at 72 weeks for the high-dose group and 104 weeks for the medium-dose group rats, alopecia, facial edema, exophthalamos, cyanosis and ambercolored urine became noticeable. Intestinal metaplasia or adenocarcinoma, were found in 3/47 (6.4%), 5/48 (10.4%), 8/48 (16.7%) or 17/48 (35.4%) of the rats in the control, medium and high dose groups, respectively. There apeared to be no differences in incidence between sexes, and incidence appeared to be dose-related.

Another laboratory (Allen et al., 1976; Allen and Abrahamson, 1979) reported on feeding diets containing 100 mg/kg diet Aroclor 1248, Aroclor 1254 or Aroclor 1262. Control rats were fed diets without added Aroclor. In all, 96 male Sprague-Dawley rats were involved for an experimental period of 52 weeks. Observations continued for 13 additional weeks. Appearance, appetite and weight gain were normal in all rats throughout the study. Hematologic parameters, (hemoglobin, hematocrit and leukocyte count) remained normal. Serum protein and albumin/globulin ratio remained normal. Total serum lipids and cholesterol remained elevated throughout the 52-week experimental period. Serum triglyceride did not vary as markedly except that rats on diets containing Aroclor 1254 had an extremely high (550% of normal) level at the 52-week sampling. Serum triglycerides from all treatment groups ranged from 20-40% below control levels, and serum cholesterol remained elevated 13 weeks after exposure to Aroclor was terminated. Changes in blood chemistries were associated with changes in the liver, which appeared as early as 13 weeks of exposure. Hepatomegaly, with livers ~1 1/2 times normal size, and focal degeneration and necrosis were evident. Hepatic protein, RNA and lipid remained increased and DNA values were decreased throughout the experimental period. Microsomal enzymes were induced and glucose-6-phosphatase was inhibited.

More intensive microscopic and ultrastructural examinations revealed considerable changes in rats exposed to 75 mg of Aroclor 1242. Jönsson et al (1981) exposed groups of six female Sprague-Dawley rats to 0, 75 or 150 mg of Aroclor 1242 for 36 weeks. Complete histological and electron microscopic examinations of the liver were performed. Upon gross examination, the livers of Aroclor 1242-exposed rats appeared much darker than those of control rats, indicative of massive venous engorgement of the liver. Histologic examination revealed areas of marked focal necrosis with moderate hepatocytic regeneration. Enlarged, vacuolated hepatocytes were commonly observed. Mitoses and multinucleate cells were common. Moderate to large amounts of pigment that stained positive for iron accumulated adjacent to central veins and, most heavily, in Kupfer cells. Upon ultrastructural examination the most striking findings were the presence of numerous pleomorphic lipid droplets in the cytoplasm. Iron deposits were present in hepatocytes, and were most prominent in the 75 mg of Aroclor 1242/kg diet group. Smooth endoplasmic reticulum exhibited marked proliferation and rough endoplasmic reticulum exhibited degranulation and degeneration. Many hepatocytes contained varying numbers of whorl-like membranous structures that were hypothesised to represent a detoxification mechanism (see the Target Organ Toxicity Section).

Chronic and Subchronic Toxicity in the Mouse -- Many of the lesions observed in rats also occurred in mice (Table V-7). Ito et al. (1973) exposed 114 male dd mice to diets containing 0, 100, 250 or 500 mg Kanechlor 300, Kanechlor 400 or Kanechlor 500 for 32 weeks. Hepatomegaly was a consistent finding in all treatment groups. Oval cell formation and bile duct proliferation occurred in the mid- and high-dose levels of Kanechlor 400 and Kanechlor 500. Mice apparently differ from rats in that they tend to develop hepatic amyloidosis as a result of exposure to PCBs. In this study hepatic amyloidosis was noted in Kanechlor-exposed mice and higher incidences were associated with the lower dietary levels of the lesser chlorinated products. Mice exposed to the high-dose levels of Kanechlor 400 and Kanechlor 500 exhibited no amyloidosis, while mice exposed to medium and low doses of these Kanechlors exhibited amyloidosis. All groups exposed to Kanechlor 300 showed amyloidosis with incidences of 8.3, 33.3 and 83.3% in the 500, 250 and 100 mg/kg diet groups, respectively. Kimbrough and Linder (1974) reported hepatopathology similar to that reported for rats and made no mention of amyloidosis. They exposed 200 male BALB/CJ mice to 0 or 300 mg Aroclor 1254/kg diet for 6 or 11 months.

Koller (1977) exposed groups of 25 male BALB/CJ mice to 0, 3.75, 37.5 or 375 mg of Aroclor 1221, 1242 or 1254/kg diet for 6 months. Observations continued for another 3 months. Aroclor 1254 resulted in the most severe manifestations of toxicity; mortality and severe hepatopathology occurred at the 375 mg/kg diet level. The middle group had fewer liver lesions, and no liver lesions were observed at the 3.75 mg/kg diet level. At 375 mg/kg diet, the Aroclor 1242-exposed mice exhibited moderate hepatopathology. Neither of the lower Aroclor 1242-exposed groups, nor any of the Aroclor 1221 groups exhibited liver lesions.

Two reports mentioned skin lesions in mice after 23 or 26 weeks of oral exposure. The first (Nishizumi, 1970) reported eczematous and ulcerative skin lesions about the head and face in mice exposed to ~1 mg of an unidentified "CBP." A more intensive study (Bell, 1983) reported skin lesions beginning with thickening and erythemia of the pinna of the ear in mice exposed to 200 mg Aroclor 1254/kg of diet.

Chronic and Subchronic Toxicity in Monkeys — One laboratory has conducted ongoing studies of the toxicity of PCBs in rhesus monkeys for several years (Table V-8). They have investigated effects of Aroclor 1248 on reproduction and fetal toxicity (see the Teratogenicity and Reproductive Toxicity Section) as well as effects of chronic exposure on adult monkeys. The initial study involved exposing 24 female and male rhesus monkeys to levels of 0, 2.5 or 5.0 mg of Aroclor 1248/kg diet for ~18 months. Observations continued for an additional 21 months (Barsotti and Allen, 1975; Allen and Barsotti, 1976; Allen et al., 1980). In this study, males appeared to be somewhat more resistant than females. Males exposed to 5.0 mg/kg diet and females at all levels of exposure exhibited palpebral edema and erythemia. Females had more severe skin lesions such as alopecia and acne. These authors suggested that the larger size of the males with their larger amounts of body fat may have provided a depot for PCBs and may have reduced apparent toxicity. Females exposed to 2.5 mg/kg diet suffered extreme weight loss and irregular menstrual cycle length with depressed serum progesterone and estradiol.

Continuing studies (Barsotti et al., 1976; Allen et al., 1979a; Barsotti, 1981) after 16 months of exposure to 2.5 mg Aroclor 1258/kg diet indicated normal hemograms. Serum chemistries were altered, total lipids were reduced, SGPT was elevated, and there was a shift in the A/G ratio after 6 months of exposure. After 12 months of exposure, serum cholesterol and triglyceride were reduced.

In another study (Allen et al., 1979b), exposure of eight female monkeys to 0.5 or 1.0 mg of Aroclor 1248/kg diet 3 times weekly for 7 months resulted in total intakes of ~8 or 16 mg of Aroclor 1248. These animals showed no irregularities of menstrual cycle or alterations in serum estrodiol or progesterone, and had normal fertility when bred. There appeared to be some fetal loss, infant birth weights were reduced and nursing infants developed focal areas of hyperpigmentation.

This laboratory subsequently exposed 24 adult female monkeys to 0.025, 0.25 or 1.0 mg Aroclor 1016/kg diet (Barsotti, 1981). Although length of time of exposure was not stated, based on the protocols of previous work performed by this laboratory indicate that exposure probably began at least 6 months before breeding and continued through 3-4 months of nursing. The author reported observing no abnormalities of clinical, gross or reproductive parameters. Infants from dams exposed to 1.0 mg/kg diet exhibited reduced birth weight.

Three surviving infant monkeys from the first study (Barsotti and Allen, 1975; Allen and Barsotti, 1976; Allen et al., 1980) were subjected to locomotor activity and learning ability tests from 6-24 months of age (Bowman and Heironomus, 1981). These subjects were infants of dams exposed to diets containing 2.5 mg Aroclor 1248/kg diet for at least 6 months before breeding through gestation and 3 months of nursing. All PCB-exposed infant monkeys exhibited hyperactivity and retarded learning ability when compared with offspring (4) of control dams. Hyperactivity continued to increase with increasing age. These same three PCB-exposed monkeys and 3 of 4 control monkeys were retested for hyperactivity at 44 months of age. At this time the three PCB-exposed monkeys showed significant ($p<0.05$) hypoactivity when compared with controls (Bowman and Heironomus, 1981).

After the original dams had been removed from Aroclor 1248-containing diets for periods of 6-18 months, they were rebred (Bowman and Heironomus, 1981). No further exposure to PCBs was allowed. Infants from these dams also showed hyperactivity when subjected to testing at 12 months of age.

Becker et al. (1979) exposed six male rhesus monkeys to levels of 3, 10, 10, 30 or 100 mg Aroclor 1242/kg diet for up to 245 days. Mortality of all monkeys exposed to ≥ 10 mg/kg diet occurred by day 245. All exposed monkeys exhibited palpebral swelling and erythemia, weight loss, rough hair coat, reduced hemoglobin and leukocytosis. Gastric lesions in exposed subjects included hypertrophic gastric mucosa consisting of elongated hyperplastic glands with destruction of parietal and zymogenic cells.

McNulty et al. (1980) also investigated the relative toxicity of 3,4,3',4'-TCB; 2,5,2',5'-TCB and Aroclor 1242. Groups of three male rhesus monkeys were exposed to 3 mg of 3,4,3',4'-TCB or 2,5,2',5'-TCB/ kg of diet or diet without added PCBs (control). The dosages were reduced to 1 mg/kg diet and again to 0.3 mg/kg diet because of rapidly developing morbidity in the three monkeys exposed to 3,4,3',4'-TCB. Mortality of two subjects in this group had occurred by day 62. Mortality of the third subject on day 215 terminated this experiment. Necropsy revealed severe emaciation with a total absence of adipose tissue. Sebaceous glands in the eyelids had undergone squamous metaplasia and become cystic. The nail beds were hyperkeratotic, nails were elevated by accumulations of keratin and many were shortened or lost. The thymus gland had undergone nearly complete regression. Gastric mucosa was severely disrupted, with development of large mucus cysts and loss of parietal and zymogenic cells. Necropsy of one control animal that was accidentally killed on day 99 and one 2,5,2',5'-TCBexposed animal intentionally killed for examination revealed no gross or microscopic lesions.

In a second experiment, groups of five young male rhesus monkeys were given diets containing 1 mg of 3,4,3',4'-TCB, 2,5,2',5'-TCB or Aroclor 1242/kg diet (McNulty et al., 1980). The control group consisted of four monkeys given food without added PCBs. At the end of the 133-day trial, all control animals, those exposed to 2,5,2',5'-TCB and those exposed to Aroclor 1242 appeared well. Biopsy samples of cheek skin taken on days 28, 63 and 114 showed normal sebaceous glands and hair follicles. In 4 of 5 monkeys exposed to 3,4,3',4'-TCB, palpebral edema and elevated fingernails were noted by the fourth week. One of these subjects died on day 33. Findings upon necropsy examination were the same as those reported in the first experiment. The remaining three sick 3,4,3',4'-TCB-exposed monkeys were placed on control diet on day 38 in an attempt to arrest their deterioration. By day 76 of recovery (day 114 of the study), examination of biopsy samples revealed normal regeneration of the skin. On day 133, the dietary level of 2, 5, 2',5'-TCB and Aroclor 1242 was raised to 5 mg/kg for an additional 2 months. All animals remained clinically well and skin morphology remained normal.

When animals from both experiments were qualitatively categorized as affected or unaffected, it appeared that 3,4,3',4'-TCB was significantly ($p<0.0001$) more toxic than 2,5,2',5'-TCB (McNulty et al., 1980). Likewise, 3,4,3',4'-TCB was ($p<0.02$) more toxic than Aroclor 1242. It is also noteworthy that nail and skin lesions occurred when rhesus monkeys were exposed to "pure" 3,4,3',4'-TCB, "free" (<1 ppm) of PCDFs.

Hori et al. (1982) investigated the toxicity of Kanechlor 400 with PCDFs removed (p-KC-400), a PCB product prepared from Kanechlor 400 containing ~400 mg PCDFs/kg (Y-PCB), and the latter product with PCDFs removed PY-PCB. Female cynomolgus monkeys were exposed to the above described sources of PCBs dissolved in salad oil and injected into 50 g pieces of banana, which were given 6 days/week for 20 weeks. Individual monkeys received 5 mg P-KC400/day, 5, 10 or 10 mg Y-PCB/day or 5 mg PY-PCB/day. Control groups consisted of two monkeys. Both 10 mg Y-PCB-exposed subjects were killed by 8 weeks because of their moribund condition. Considerable weight loss (13 and 20%) occurred in the 5 mg P-KC-400 and Y-PCB-exposed monkeys, respectively. Monkeys exposed to 5 or 10 mg Y-PCB exhibited typical skin lesions that included partial to complete alopecia, acne, hyperpigmentation, periorbital edema and dermal hyperkeratosis. Other clinical signs in the P-KC-400 monkey were mild or not apparent.

Both P-KC-400, Y-PCB and PY-PCB-exposed monkeys exhibited depressed immunocompetency as evaluated by titer developed against sheep red blood cells (Hori et al., 1982). There appeared to be no differences between P-KC-400 and Y-PCB-exposed monkeys in their immune response. Histopathological examination of the P-KC-400, Y-PCB and PY-PCB-exposed monkeys yielded similar lesions. The livers showed areas of focal necrosis and hepatocytic enlargement with increased smooth endoplasmic reticulum. Proliferation of bile duct cells was another finding. Kidneys were enlarged because of dilated renal tubules. The tubular epithelial cells were enlarged and vacuolated. Kidney tubules contained cylindrical casts. Lesions were more severe in the Y-PCB-exposed monkey. The formation of meibomian gland cysts was seen only in the Y-PCB-exposed monkey. Hori et al. (1982) concluded that PCDFs are more toxic than PCBs, particularly to the skin.

Chronic and Subchronic Toxicity in Other Species -- Chronic and subchronic toxicity in other species are summarized in Table V-9. Mink have been determined to be particularly sensitive to PCBs, with respect to reproductive performance (see the Teratogenicity and Reproductive Toxicity Section). Waste from plants that process Great Lakes fish has been used as an economical source of food for mink on fur farms. Early in the 1960s, complaints arose from those feeding Great Lakes fish that reproductive success was considerably decreased. PCBs were suspected and one laboratory

(Aulerich et al., 1973; Aulerich and Ringer, 1977; Bleavins et al., 1980) investigated this phenomenon in detail. In one study (Bleavins et al., 1980) Aroclor 1242 and 1016 were fed to 105 male and female pastel mink for ~8 months. Aroclor 1242 was fed at 0, 5, 10, 20 or 40 mg/kg diet and Aroclor 1016 was fed at 0 or 20 mg/kg diet. Aroclor 1242 at levels ≥20 mg/kg diet resulted in 100% mortality. Aroclor 1016 at 20 mg/kg diet resulted in mortality of 25% (3/12) of the females exposed. Necropsy revealed emaciation, an almost complete absence of body fat and gastric ulceration. Reproductive performance was reduced by 20 mg Aroclor 1016/kg diet and completely eliminated by ≥5 mg Aroclor 1242/kg diet.

An earlier study (Aulerich and Ringer, 1977) indicated that 2 mg/kg of Aroclor 1016, 1221, 1242 or 1254/kg diet had no effect on body weight gain, hemoglobin or hematocrit in minks. Aroclor 1254 at 2 mg/kg diet caused severely decreased reproductive performance.

One study evaluated the relative toxicity of Aroclor 1221, 1242 and 1254 in rabbits (Koller and Zinkl, 1973). Groups of five male and five female New Zealand white rabbits were treated by gavage with 300 mg of each Aroclor once weekly for 14 weeks. A control group consisting of five males and five females received corn oil alone. Aroclor 1254 proved to be the most toxic product tested, causing a reduced rate of weight gain, severe hepatomegaly and uterine atrophy. Hepatocytes were enlarged, vacuolated and undergoing granular degeneration. Areas of considerable centrilobular necrosis and fibrous organization were noted. Liver lesions were much less severe in the Aroclor 1242-exposed groups and absent in the Aroclor-1221 exposed groups. Both Aroclor 1242 and 1254 caused elevated SGOT and SGPT. Aroclor 1254 caused a transient increase in serum cholesterol after 7 weeks of exposure.

Hematologic parameters, BUN and serum protein fractions were unaffected by treatment. Based on their findings, these authors concluded that the more heavily chlorine-substituted PCBs were more toxic to rabbits than were less heavily chlorinated PCBs.

Target Organ Toxicity

Hepatic Effects. Hepatoxicity caused by exposure to PCBs is discussed in practically every paper that deals with PCB toxicity. Clearly, the liver is the organ most often implicated in toxicity of PCBs. Several reviews mentioned at the beginning of this section are recommended for a general discussion of PCB-induced hepatotoxicity. This section will not, therefore, be exhaustive, but will attempt to focus on those studies that elucidate particularly well changes in morphology, ultrastructure, biochemistry and detoxification in the liver.

Although hepatopathology has been reported in all species studied, the more detailed reports of morphological and biochemical alteration deal with the rat. For that reason, most of the studies discussed have involved the rat. One of the earlier reports on microscopic and ultrastructural changes in the liver associated with PCBs was made by Kimbrough et al. (1972). The authors exposed groups of 10 Sherman rats of each sex to 20, 100, 500 or 1000 mg Aroclor 1260/kg diet or 20, 100 or 500 mg Aroclor 1254/kg diet for 8 months. In this study Aroclor 1260 appeared to be more toxic to females rather than male rats, because two of the 500 mg/kg diet-exposed and eight of the 1000 mg/kg diet-exposed groups died. None of the control rats or Aroclor 1260-exposed males died. Upon necropsy, livers of males at all levels of exposure were significantly larger than those of controls. Livers of females showed a small but insignificant trend toward hepatomegaly.

Porphyria, evidenced by UV fluorescence, occurred in many rats. Fluorescence was most pronounced in the liver, with occasional fluorescence of bone, serum or urine or both. Grossly, livers were soft and varied from yellowish-brown to dark olive. Grayish white nodules up to 0.5 cm in diameter were seen in a number of livers.

Microscopically, hypertrophy of individual liver cells was a constant finding (Kimbrough et al., 1972). Nuclei were often pleomorphic and hyperchromatic; occasional mitoses were observed. The cytoplasm either contained large vacuoles or appeared foamy. Kupfer cells and perivascular macrophages contained brown pigment that stained iron-positive with Perls' stain. Areas of adenofibrosis were found which correspond to the grayish-white nodules described grossly. Ultrastructurally, an increase in the smooth endoplasmic reticulum occurred in the hepatocytes of all exposed rats. Large and small lipid-containing vacuoles were present. Occasionally atypical mitochondria were observed. At dietary levels of 500 and 1000 mg/kg, concentrically arranged membranes were seen to surround lipid-containing vacuoles. At the lower dietary levels dark and light cells, granular cytoplasmic inclusions and dilated bile ducts were noted. Widening of intracellular spaces, increased collagen and occasional phagocytosed erythrocytes were observed. Areas of adenofibrosis consisted of fibroblasts with large amounts of collagen surrounding rosettes of epithelial cells. Some of these bile duct cells contained mucus and resembled goblet cells. Examination of livers from Aroclor 1254-exposed rats resembled those from Aroclor 1260-treated rats. Aroclor 1254 seemed to result in a much higher incidence of adenofibrosis at lower dietary levels. As a result of observed mortality, the authors concluded that the more heavily chlorinated the PCBs were, the more toxic they were. On the other hand, Aroclor 1254 seemed to result in adenofibrosis at lower dietary levels than did Aroclor 1260, suggesting that the Aroclor product containing the lesser amount of chlorine may be more hepatotoxic, at least in the induction of adenofibrosis.

Ito et al. (1973) exposed 114 male dd mice to levels of 100, 250 or 500 mg Kanechlor 300, Kanechlor 400 or Kanechlor 500/kg diet for 32 weeks. They reported hepatomegaly, oval cell formation and bile duct formation with increasing incidence apparently related to both amount and degree of chlorination of the Kanechlors. An interesting finding was the presence of amyloidosis, which occurred with a greater incidence when smaller doses of less heavily chlorinated Kanechlors were fed in the diet. No explanation of this phenomenon or its significance was offered.

In a study investigating the hepatotoxicity of various "pure" PCB isomers, Hansell and Ecobichon (1974) administered 50 mg of the test compounds/kg bw by i.p. injection. Groups of seven young male Wistar rats were treated daily for 3 days. Compounds tested were Aroclor 1254; Aroclor 1260; biphenyl; 4-MCB; 2,2'-DCB; 2,4'-DCB; 4,4'-DCB; 2,5,2',5'TCB; 2,4,2',4'-TCB; 3,4,3',4'-TCB; 2,4,5,2',4',5'-HCB; 2,4,6,2',4',6'-HCB; 2,3,5,2',3',5'-HCB and 2,3,4,5,2',3',-4',5'-OCB. Significant increases in liver weight occurred following exposure to Aroclor 1254, Aroclor 1260 and 2,3,5,2'3'5'-HCB. The hexa- and octa-chlorinated biphenyls produced alterations detectable by conventional histopathological procedures; these included hepatocytes with vacuolated cytoplasm and focal necrosis involving 5-6 cells. A subjective evaluation of proliferation of smooth endoplasmic reticulum, cytoplasmic formation of lipid droplets and increased presence of "microbodies" was made to indicate relative toxicity of compounds tested. Based on these three ultrastructural parameters, Aroclor 1254 appeared to cause greater hepatic alteration than Aroclor 1260 and seemed to be the most toxic material tested. The OCB and HCBs tested appeared to be uniform in effects on hepatocytes and, collectively, seemed to be more toxic than TCBs, DCBs or MCB. Of the TCBs, 2,4,2',4'-TCB appeared to be the most toxic. The 2,4'- or 4,4'-DCB appeared to be equally toxic; both seemed more toxic than 2,2'-DCB.

In a second experiment 4-MCB 4,4'-DCB and 2,5,2',5'-TCB were administered to rat by i.p. injection at a daily dose of 100 mg/kg bw for 7 days (Hansell and Ecobichon, 1974). The 4,4'-DCB isomer caused more dramatic alterations than either of the other compounds tested. The authors reported the presence of concentric membranes or whorls in the cytoplasm, which often contained lipid droplets. These whorls may originate from rearrangement of rough endoplasmic reticulum, and may represent a means by which lipid droplets containing lipophilic toxins are segregated from the rest of the cell.

Kasza et al. (1978b) exposed groups of seven 4-week-old male Holtzman rats to 0, 5, 50 or 500 mg Aroclor 1254/kg diet for 5 weeks. They observed hepatic alterations practically identical to those previously described in this section. Additionally, they reported a dramatic and dose-related increase in the number of liposomes, as well as lipid droplets. They also reported a considerable increase in the number of Golgi apparatus at the 5 mg/kg diet exposure level and a modest reduction in Golgi apparatus in the 50 and 500 mg/kg-exposed groups. Laminated cytoplasmic inclusions or membrane whorls were seen in hepatocytes from rats exposed to 500 mg/kg diet.

These were thought to arise from hypertrophic smooth endoplasmic reticulum, and to represent a normal stage in membrane formation or repair. The authors reported an accumulation of liposomes concomitant with an initial increase in Golgi apparatus associated with condensing vesicles containing lipoprotein. This suggested that a blockage occurred in the mechanism by which hepatocytes discharge lipids. This process apparently involves conjugation of protein and lipid, transport by the endoplasmic reticulum and Golgi apparatus, intracellular migration through microtubules and secretion into the intracellular space.

Recently, Jonsson et al. (1981) presented evidence that led them to strongly hypothesize a detoxification role for the membranous whorls described earlier. They exposed mature female Sprague-Dawley rats to diets containing 75 or 150 mg Aroclor 1242/kg for 8 or 36 weeks. There were six animals in each group. Ultrastructural features of livers from PCBexposed rats were similar to those reported from the other studies in this section. Numerous pleomorphic lipid inclusions dominated the hepatocytic cytoplasm. Many of the lipid droplets contained clear, punched-out areas suggestive of the presence of lipid-cholesterol complexes as might be seen in normal adrenal cortical cells. Smooth endoplasmic reticulum had proliferated, and rough endoplasmic reticulum had undergone degranulation and degeneration. Many hepatocytes in all experimental groups exhibited varying numbers of whorl-like membranous bodies. Some were found within the cytoplasm, some near the periphery and some free in the intercellular spaces. Because an occasional whorl was "caught" undergoing exocytosis, the authors hypothesized that this phenomenon represented a hepatocytic detoxification mechanism.

Koller and Zinkl (1973) administered 0 or 300 mg Aroclor 1221, 1242 or 1254 by stomach tube, once weekly for 14 weeks, to New Zealand White rabbits. Blood samples were taken every 2 weeks from the five males and five females in each group for determination of blood chemistries. After 2 weeks SGPT and SGOT levels were elevated in Aroclor 1254-treated males. Females developed elevated SGPT and SGOT at 4 and 8 weeks, respectively, after exposure. Aroclor 1221 failed to elevate serum levels of either enzyme throughout the study. Total serum protein, protein fraction levels and BUN did not differ from controls during the study. No hematologic differences were noted between control and treatment individuals. Serum cholesterol was elevated significantly in males treated with Aroclor 1254 as early as 7 weeks. The authors concluded that Aroclor 1254 is more toxic to rabbits than Aroclor 1242, and that Aroclor 1221 appeared to be relatively nontoxic in this species. Elevated SGPT, SGOT and serum cholesterol were interpreted as evidence of hepatotoxicity. The authors examined the livers of all animals at the end of the 14-week study. Ultrastructural alterations were found in livers from Aroclor 1254- and, less well-developed, from Aroclor 1242-treated animals. Livers from Aroclor 1221-exposed animals did not differ from those of controls.

A one-time i.p. administration of 100 mg Clophen A-50/kg bw to male Wistar rats elicited profound biochemical changes in the liver (Parkki et al., 1977). Cytochrome P-450 levels increased 3- to 4-fold with a maximum achieved in 1 week. NADPH activity doubled, and p-nitroanisole-o-demethylase activity increased 6- to 7-fold and declined ~4 times normal activity at the end of the 4-week observation period. AHH activity increased 3-fold initially and had returned to normal by the end of 4 weeks. Microsomal expoxide hydratase activity had increased 2.5-fold at 1 week and persisted at this level. Glutathione-S-transferase activity had increased by 1 day and remained elevated for 4 weeks. This study pointed out the rapidity with which certain hepatic exzyme activities are induced and their persistence in the induced form.

Allen and Abrahamson (1973) fed diets containing 0 or 1000 mg Aroclor 1248, 1254 or 1262/kg to male Sprague-Dawley rats for 6 weeks. Examinations of 4 rats from each group were performed on days 1, 3, 7, 14, 21, 28 and at the end of the 6-week trial. They reported reductions in weight gain, most noticeable in Aroclor 1248- and least noticeable in Aroclor 1262-exposed rats. Liver weights at the end of 6 weeks were increased >3-fold, with livers from the Aroclor 1262-exposed rats most enlarged and livers from Aroclor 1248-exposed rats least enlarged. Initially, all Aroclors tested resulted in an increase in liver protein and RNA, with Aroclor 1262 being most effective and Aroclor 1248 being least effective. At the end of 6 weeks, phospholipid and cholesterol were elevated, most highly in livers from Aroclor 1248-exposed and least from Aroclor 1262-exposed rats.

In this study, many enzyme systems were induced by 1 week of exposure to all of the Aroclors tested (Allen and Abrahamson, 1973). Microsomal nitroreductase activity increased initially, and declined to near normal values by the second week. Activity continued to decline in following weeks except in livers from Aroclor 1260-exposed rats, in which activity remained high. N-demethylase activity in hepatic microsomes from PCB-exposed rats persisted at high levels throughout the 6-week period. Aromatic hydroxylase, glucose 6-phosphatase and esterase activity began to decrease almost immediately, and continued a downward trend throughout the study.

Ultrastructural studies on the livers were conducted simultaneously with the compositional biochemical determinations (Allen and Abrahamson, 1973). The authors noted the tremendous enlargement of endoplasmic reticulum mentioned earlier in this section, and correlated the occurrence of enzyme induction with this phenomenon. By 6 weeks, degenerative changes such as vesiculation of the endoplasmic reticulum, dissolution of membranous whorls and accumulation of numerous lipid droplets had occurred. This degeneration was accompanied by a decrease in hepatic enzyme activities. The authors therefore suggested that exposure to PCBs resulted in an initial hepatic induction phase followed by degeneration when exposure was too high (100 mg/kg diet) levels. Although greater hepatic hypertrophy was associated with the more chlorinated Aroclors, greater toxicity seemed to result from less highly chlorinated Aroclors.

Allen and Abrahamson (1979) exposed male Sprague-Dawley rats to diets containing 0 or 100 mg Aroclor 1248, 1254 or 1262/kg for 1 year. Partial (70%) hepatectomy was performed on four rats from each group at the end of 13, 26, 39 and 52 weeks of exposure. Survivors at the end of 52 weeks were put on a control diet for 13 weeks before being killed for final examination. Ratios of hepatic protein/DNA and RNA/DNA remained fairly uniformly elevated throughout the 52-week exposure period to any of the three Aroclors tested. Levels were returning to near control levels by 65 weeks. Aroclor 1248 seemed less capable of maintaining elevated protein/DNA or RNA/DNA ratios than the more highly chlorinated Aroclors tested. Liver lipids increased constantly during the 52-week exposure period but had decreased substantially toward control levels after 13 weeks of control diet. Aroclor 1248 had resulted in highest lipid content by 52 weeks of exposure and

Aroclor 1263 had resulted in the least, substantiating the authors' previous claims that the less chlorinated Aroclors were the more toxic (Allen and Abrahamson, 1973). Throughout the 65-week study period N-demethylase and nitroreductase activities remained elevated. Glucose 6-phosphatase activity remained uniformly reduced. The authors concluded that hepatic microsomal functions can be induced and maintained at fairly high levels for a year or more when "moderate" dietary levels of PCBs are fed to rats.

Immunological Effects. Dermal applications of 120 mg of either 2,4,5,2',4',5'-HCB or Aroclor 1260, 5 times weekly for 4 weeks resulted in moderate thymic atrophy in rabbits (Vos and Notenboom-Ram, 1972). The administered dosages of these test substances were sufficient to cause serious liver lesions, with the more severe lesions found in the 2,4,5,2',4',5'-HCB-exposed animals (Table V-4). Both test substances resulted in similar (moderate thymic atrophy, indicating equal potential for cell-mediated immunosuppression.

Allen and Abrahamson (1973) (Table V-2) observed a lymphocytopenia, relative neutrophilia and decreased thymic weight in male Sprague-Dawley rats exposed to Aroclors 1248, 1254 or 1262. Dietary levels of 0 or 1000 mg/kg were fed for up to 6 weeks.

A single i.p. administration of 1000 mg Aroclor 1242/kg bw to BALB/CJ mice resulted in splenomegaly (Carter and Clancey, 1980) (Table V-4). Splenomegaly was significant by day 6, had peaked by day 9 and had regressed by day 13. Examination of the cellular components revealed a significant reduction in lymphocytes on days 6-10.

Recently, Biocca et al. (1981) exposed groups of five male C57Bl/6J mice to four symmetrical HCB isomers to evaluate their relative toxicity (Table V-3). Dietary levels of 0.3, 1.0, 3, 10, 30, 100 or 300 mg/kg were used for 28 days. It appeared that 3,4,5,3',4',5'-HCB was the most toxic isomer tested. As little as 3.0 mg/kg resulted in moderate thymic involution; 30 mg/kg caused marked involutions. As little as 3.0 mg/kg diet caused moderate depletion of splenic lymphocytes.

It appeared that levels of PCBs that caused histological alterations in other organs, notably the liver, and frequently caused minimal alterations in thymus and spleen. It is possible, however, that other parameters, such as reduced production of antibodies or increased mortality to challenge with endotoxin or live organisms, may indicate toxicity at lower exposure levels.

Vos and van Genderen (1973) exposed 40 male guinea pigs to 10, 50 or 250 mg Clophen A-60/kg diet for 4 weeks. Hemaglutination titers to a single dose of tetanus toxoid injected in the foot pad were significantly reduced at the 50 mg/kg level, and it appeared that the response was doserelated from the 10 mg/kg diet level. Tetanus antitoxin producing cells in the isolateral popliteal lymph node showed a substantial reduction in animals exposed to 250 mg/kg diet. Female guinea pigs exposed to 50 mg Clophen A-60 or Aroclor 1260/kg diet for 6 weeks showed significant ($p<0.025$) decreases in both total white blood cell and lymphocyte levels. Single radial immunodiffusion testing revealed reduced tetanus antitoxin titers following injection with two doses of tetanus toxoid.

To measure the effects of PCBs on cell-mediated immunity, 30 female guinea pigs were fed diets containing 50 or 250 mg Clophen A-60 for 7 weeks and stimulated with Freund's Complete Adjuvant. Skin reaction after intradermal tuberculin administration was measured at 24 and 48 hours. Total leukocytes, but not lymphocytes, were reduced significantly by exposure at ≥50 mg/kg. Both total leukocyte and lymphocyte levels were low in control animals; no explanation was offered. Thymus weights were significantly reduced ($p<0.05$) as was the skin tuberculin reaction ($p<0.005$) at both 24 and 48 hours.

The most extensive studies of PCB-induced immunosuppression were performed in mice (Loose et al., 1978a,b; Silkworth and Loose, 1978). BALB/CJ mice were used in all trials measuring survival following administration of endotoxin or noxious organisms. C57Bl/6 mice were used to evaluate the graft vs. host response. Diets containing 167 mg of Aroclor 1242 or Aroclor 1016/kg were fed for 3 or 6 weeks. In the first trial, a diet containing Aroclor 1242 was fed for 6 weeks to groups of ~13 mice (Loose et al., 1978a). _Salmonella typhosa_ endotoxin administered at this time resulted in increased ($p<0.05$) mortality in treated mice. Another group of mice similarly treated were injected with malarial organisms, _Plasmodium berghei_. Again, survival was significantly ($p<0.05$) reduced at the end of 3 weeks in treated vs. control mice.

In a companion study, the numbers of primary and secondary splenic plaque-forming cells in response to sheep erythrocytes was estimated (Loose et al., 1978a). Exposure to 167 mg Aroclor 1242/kg diet for 6 weeks resulted in a >2-fold reduction in splenic plaque-forming cells. In this study both memory cell function and serum IgA levels were depressed when mice were immunized with sheep erythrocytes 3 weeks before PCB exposure occurred. Subsequently, the authors exposed mice to 167 mg Aroclor 1242 or Aroclor 1016/kg diet for 6 weeks. Challenge with _S. typhosa_ endotoxin or _P. berghei_ organisms resulted in increased ($p<0.05$) mortality in PCB-exposed mice. There were no differences in responses in mice exposed to the two different Aroclors. The authors stressed that these demonstrations of suppressed immunocompetency occurred in the absence of detectable histopathologic lesions in lung, thymus, mesenteric lymph nodes or spleen. The liver was also examined histopathologically, but hepatocytic hyperplasia was the only lesion reported.

In a short abstract, Silkworth and Loose (1978) reported that Aroclor 1016 elicited a greater graft vs. host response when splenic cells from treated mice were injected into neonates. Splenic cells were harvested from mice exposed to 167 Aroclor 1016/kg diet for 3 weeks.

Thyroid Effects. The thyroid gland was selected for discussion as a target organ primarily because of the work of Collins et al. (1977). Their work clearly delineated the effects of PCBs on the thyroid and suggested how depressed thyroid levels may relate to some of the other symptoms of PCB toxicity. In their first study, Collins et al. (1977) exposed 8-week-old male Osborne-Mendel rats to 50 or 500 mg Aroclor 1254/kg of diet for 4 or 12 weeks. Animals on 500 mg/kg diet were reduced to 250 mg/kg diet at 6 weeks because of anorexia and weight loss. Thyroid glands were collected from 10 rats from each treatment level at 4 weeks and 12 weeks of treatment, followed by 12 or 35 weeks of recovery. Blood was collected from five rats at each dose and time interval for the thyroxine assay.

After 4 weeks on 500 mg/kg diet, the thyroid glands of rats were enlarged and composed of small, irregularly-shaped follicles (Collins et al., 1977). In some areas, columnar cells were arranged in multiple layers. Follicular cells were hyperplastic, with papillary projections extending into the lumens. Numerous cytoplasmic processes projected into the colloid from the apical surfaces of follicular cells. Acid phosphatase activity was increased in the cytoplasm of follicular cells. Thyroids of rats fed 50 mg/kg diet for 4 weeks had similar but milder lesions. By 12 weeks of exposure to either the low or high levels of Aroclor 1254, the cytoplasm of thyroid follicular cells had become vacuolated, and the nuclei were basically situated. Follicular cells had become more columnar, and the rough endoplasmic reticulum appeared irregularly dilated with finely granulated material. The Golgi apparatus was compressed near the nucleus and was less extensive than in control rats. Mitochondria were enlarged and frequently swollen, with disrupted cristae. Large numbers of lysosomal bodies and colloid droplets were present in the cytoplasm. Microvilli on the luminal surfaces of follicular cells appeared to be abnormally short, blunt and branched. Extensive areas of the luminal surfaces of follicular cells were devoid of microvilli and occasional unique cytoplasmic projections extended into the colloid. Acid phosphatase activity was very strong. Serum thyroxine levels over this period decreased from 6.66 and 7.18 μg/dℓ in controls to 4.8 and 1.96, and 2.10 and 1.78 μg/dℓ in the 50 mg and 500 mg/kg diet groups over the 4- and 12-week exposure periods, respectively ($p<0.005$).

The thyroid gland appears to have remarkable capabilities of regeneration. By 5 weeks of recovery thyroid follicles were again composed of single layers of columnar cells that were only occasionally vacuolated (Collins et al., 1977). By 12 weeks of recovery, thyroid glands of treated rats resembled those of controls. Thyroid follicles were lined by a single layer of colloidal or low columnar cells with centrally located nuclei. The endoplasmic reticulum appeared to be well-developed and consisted of long profiles with narrow cisternae. Few areas of dilated cisternae remained. The Golgi apparatus had regained its prominence and was associated with numerous dense granules. Mitochondria had returned to normal with only occasional swelling and disruption of cristae. Lysosomal bodies and colloid droplets were decreased to numbers found in thyroids of control rats. By 12 weeks, serum thyroxine levels had recovered to ~50% of normal levels. By 35 weeks, serum thyroxine recovery was complete.

The progressive accumulation of large lysosomes with strong acid phosphatase activity, mitochondrial disruption, altered follicular cell microvilli and cytoplasmic projection into luminal colloid were interpreted to be a direct effect of PCBs on the thyroid gland (Collins et al., 1977). These changes were associated with a significant decrease in serum thyroxine after 4 weeks. Hypertrophy of follicular cells with dilated endoplasmic reticulum suggested a compensatory attempt on the part of the cell to increase secretory activity. It appeared that the increased number of lysosomal bodies with strong acid phosphatase activity were incapable of causing hydrolysis of the colloid thyroglobulin and release of active thyroid hormone, resulting in depressed levels of circulating thyroxine. Additionally, it has been shown that PCBs enhance the peripheral metabolism of thyroxine (Bastomsky, 1974), which would further reduce circulating thyroxine (Chapter VII).

On the basis of these results, Collins et al. (1977) hypothesized that some of the other symptoms associated with PCB intoxication may be due to altered thyroid function. They included decreased weight gain, reduced rate of gain, decreased reproductive performance and dermal hyperpigmentation among the symptoms that were possibly related to reduced levels of thyroxine.

In a later study with 4-week-old Holtzman rats, Kasza et al. (1978a) fed diets containing 5, 50 or 500 mg Aroclor 1254/kg diet for 5 weeks. At the end of 5 weeks, the rats were killed and thyroid glands were examined. At the lower level of 5 mg/kg diet (0.25 mg/kg bw), thyroid glands resembled histologically those of control rats. Both the colloid and follicular cells were only moderately vacuolated. Ultrastructurally, at the 5 mg/kg diet level, follicular cells were more columnar and the endoplasmic reticulum showed considerable dilatation. Abnormal lysosomes and colloid droplets accumulated in follicular cell cytoplasm. Microvilli of the follicular cells were shortened, blunted and occasionally abnormally branched. Unfortunately, serum thyroxine levels were not assayed. The authors concluded that levels as low as 5 mg Aroclor 1254/kg diet for 5 weeks resulted in significant (but probably reversible) ultrastructural and functional changes in the thyroid gland.

Subsequently, Collins and Capen (1980a) exposed homozygous and heterozygous Gunn rats, 6/group, to 0 or 500 mg Aroclor 1254/kg diet for 42 days. Homozygous Gunn rats are known to have an impaired ability to conjugate thyroxine with glucuronic acid, and hence elimination is reduced. Retro-orbital blood was collected for serum thyroxine determinations. Labeled ^{125}I-thyroxine was injected i.v. and the ratio of biliary:plasma ^{125}I-thyroxine was determined 18 hours later. Both homozygous and heterozygous Gunn rats experienced ultrastructural changes in the thyroid gland similar to those described earlier (Collins et al., 1977; Kasza et al., 1978a) with the more progressive changes occurring in the heterozygous rats. Serum thyroxine levels of control heterozygous and homozygous rats were similar. Serum thyroxine levels of PCB-exposed heterozygous and homozygous rats were similar but reduced to ~21-30% of the levels of their respective controls. Bile:plasma ratios of labeled ^{125}I-thyroxine were markedly elevated (~4-fold) in heterozygous rats exposed to PCBs when compared with heterozygous controls. Homozygous rats exposed to PCBs had about a 2-fold increase in bile:plasma ratios of ^{125}I-thyroxine. These data suggested that PCBs resulted in a lesser degree of increased thyroxine hepatic conjugation and excretion in homozygous rather than heterozygous Gunn rats. The authors concluded that both decreased thyroxine secretion and increased hepatic metabolism accounted for decreased thyroxine levels in PCB-exposed rats.

Carcinogenic, Mutagenic and Reproductive Effects

Carcinogenicity. PCBs have been tested for carcinogenicity in rats and in mice by incorporation of particular commercial PCB preparations into the diet. Six separate PCB feeding studies and one study of topical application to the skin are considered. Studies in which the PCB preparations were administered in conjunction with another agent are also discussed in this section.

The feeding studies demonstrate the carcinogenicity of some commercial PCB preparations although it is not known which of the PCB congeners in such preparations, or which of their metabolites, are responsible for the carcinogenicity demonstrated by the tests.

The liver appears to be the primary target organ which exhibits a tumorigenic and carcinogenic response to PCB exposure. The studies reviewed had, in varying degree, shortcomings which modify the meaning of the results and the contribution the study made to the overall assessment of PCB carcinogenicity. The significance of the presence, absence and severity of the liver lesions cannot be taken at face value, as if all experiments and their results had equal weight. Critical technical assessment of the design, execution and data are required for each experiment before valid conclusions can be drawn. Such critical assessments follow. Conclusions concerning the matter of PCB carcinogenicity, based on these technical evaluations, are found at the end of the section.

The feeding studies on rats are considered first, followed by the feeding studies on mice. Studies by other routes of administration and which employed more than one agent are considered after the feeding studies.

Rat Studies -- Kimbrough et al. (1975) found that Aroclor 1260 unequivocally produced hepatocellular carcinoma in female Sherman rats when 100 ppm dietary level was administered for 21 months (630 days). In this study, complete histophathology on all major organs was carried out on 200 treated rats. There was a slight but statistically significant decrease in weight gain after 3 months in the treated group. The average final body weight of treated animals was 392 g while that of controls was 420 g. The difference in weight gain was not large enough to consider the dose level to have exceeded the MTD. There were no other definite dose-related signs of toxicity.

Hepatocellular carcinoma was found in only 0.58% of the controls (1/173) and in 14% (26/184) of the treated animals. In addition, there were 0/173 control animals with neoplastic liver nodules as compared to 78% (144/184) of the treated animals. All in all, there was less than 1% incidence of neoplastic liver lesions in controls as compared to 92% (170/184) of such lesions in treated animals. Although only a single dose was selected and only female animals employed, the study demonstrates hepatocarcinogenicity of Aroclor 1260 in female Sherman rats. It is of interest to note that Kimbrough et al. (1972) in a preliminary experiment using 10 animals of each sex for each of three doses (100, 500 and 1000 ppm) of Aroclors 1260 and 1254, did not produce either neoplastic nodules or hepatocellular carcinoma in this same strain of rat when the study ran less than a year. In this preliminary study Aroclor 1254 produced adenofibrosis in 10/10 male rats. This finding suggests that hepatocellular carcinoma results when the dose is low enough to permit the study to be run for a long enough time without interfering toxicity. The 14% incidence of hepatocellular carcinoma in the large experiment also explains why it would be unlikely to have detected this cancer in experiments run a on small number of animals. Fourteen percent of 24 animals would only be 3-4 animals and they would only have appeared after about a year. Experiments which reduced the number below 24 before the earliest time to tumor would not be expected to yield a detectable carcinoma incidence.

We note also that although this experiment is probably the best rat study to use for risk assessment, it also has a problem in that the tumor incidence produced by the mixture is the result of a mixture of molecules each capable of contributing or failing to contribute to the carcinogenic process. The active ingredient(s) in the mixture most likely is limited to a few of the molecular species in the mixture. A tumor yield of 14% due to the presence of a molecular species which comprises only a fraction of the composition of the administered material would be very potent indeed.

Kimura and Baba (1973) fed diets containing Kanechlor 400 to groups of 10 male and 10 female Donryu rats. Controls consisted of five rats of each sex. Kanechlor 400 was incorporated into the test diet initially at 38.5 ppm and increased periodically to an upper limit of 616 ppm to keep pace with body weight gain. The upper level resulted in severe body weight loss and was accordingly reduced to 462 ppm for the remainder of the trial. The duration of the study ranged from 159 days to 538 days for the different animals, but there were two intervals of 4 weeks each during which the Kanechlor feeding was stopped so that the total feeding of Kanechlor was ~400 days. Female rats consumed 700-1500 mg and male rats consumed 450-1800 mg Kanechlor 400 by the end of the trial.

Adenomatous nodules in the liver were found in 6/10 female rats which had consumed 1200 mg or more of the Kanechlor 400. The remaining females that had received less than 1200 mg Kanechlor 400 and all of the males (even those that received more than 1200 mg) did not have such lesions. All treated animals, of both sexes, showed fatty degeneration of the liver.

It is important in evaluating this study to consider the weight data supplied by the investigators. These data were sufficient to determine that the treated animals showed weight decrements of >20%. Such decreases in weight gain may indicate that the MTD was exceeded.

The animal weights record of the experiment showed the average initial weight for control males was 205 g and the final weight was 462 g. In contrast, males which had received 1200 mg Kanechlor 400 or more started with an average weight of 210 g and had a final average weight of 259 g. Among female controls the initial average weight was 196 g and the final weight average was 323 g. Among those 6 treated females which had received 1200 mg or more PCB, the initial weight was 196 g and their final weight was still only 196.0. Table V-10 shows the percentage of weight gain for various doses and for controls.

It is clear from these weight data that in all cases the treated animals received doses of Kanechlor 400 which exceeded the MTD as judged by current criteria. Toxic symptoms not only included failure of rats to gain weight at a normal rate but 80% of the females and 10% of the males showed depilation; 70% of the females and 60% of the males had lung abscesses; 30% of the females and 10% of the males had intracranial abscesses; fatty degeneration of the adrenal was also found in treated animals. These lesions were not reported in the control animals. The lesions and toxic manifestations found after 400 days of treatment would have undoubtedly been even worse had the experiment been carried out for the usual two-year duration of a carcinogenicity study. The relationship between chronic toxic manifestations in these animals and liver tumorigenicity is difficult to determine. Evidently liver toxicity is a major effect of high doses of Kanechlor 400.

The study was too short and the exposure level too high to provide a good experimental basis for the determination of the carcinogenic potential of the PCB preparation used.

TABLE V-10

Weight Data; Further Analysis of Changes*

PCB in Diet (ppm)	No. Animals	Initial Weight (g)	Final Weight (g)	Percent Difference
		MALES		
450	1	199	309	+55
900	1	192	273	+42
>1200	8	210	259	+23
Control	5	205	462	+125
		FEMALES		
700	1	158	137	-13
1100	3	160	162	0
>1200	6	196	196	0
Control	5	196	323	+65

*Source: Kimura and Baba, 1973

Kanechlor 300, Kanechlor 400 and Kanechlor 500 were studied in male Wistar rats by administration of 100, 500 or 1000 ppm of each of the test preparations in the diet. The treatments lasted for 1 year in the case of Kanechlor 500 and Kanechlor 300, but for only 28 weeks for the Kanechlor 400 at the 500 ppm level and for 40 weeks at the 100 and 1000 ppm levels.

The initial weights for each group and their respective weight gains are given in Table V-11.

The figures indicate that the weight gain was inversely related to the dose of Kanechlor used for the 40 and 52 week exposure groups. In all cases (for 52 week exposure) the average percentage weight gain was below that observed with controls, but only marginally below controls for the lowest dose groups of 100 ppm. The weight figures indicate that at 100 ppm for 52 weeks, Kanechlors 300 and 500 did not produce a toxic manifestation represented in weight loss. At the 100 ppm level the liver weights were (expressed as percentage of body weight) 2.4% for controls, 2.9% for the Kanechlor 300 and 3.7% for the Kanechlor 500. There was no amyloidosis, cholangiofibrosis or fibrosis at 100 ppm for these two Kanechlors. At higher doses oval cell proliferation, bile duct proliferation, fatty changes and cellular hypertrophy were seen. Since the 100 ppm level did not seem to produce other toxic manifestations the finding of nodular hyperplasia, which the investigators considered to be preneoplastic, in 3/25 animals treated with Kanechlor 500 and 1/22 animals treated with Kanechlor 300 is noteworthy. The study does not provide data on the progression of preneoplastic to neoplastic effects since it was terminated after 52 weeks.

TABLE V-11

Percentage Average Weight Increased Calculated*

Product	ppm	Initial Weight Gain Average	Percent Increased Average	No. Animals
		52-Week Exposure Cases		
K-500	1000	126	129	13
	500	123	207	16
	100	124	300	25
K-300	1000	128	228	15
	500	135	239	19
	100	125	304	22
Control	0	130	325	18
		40-Week Exposure Cases		
K-400	1000	163	23	10
	100	175	129	16
Control	0	--	--	--
		28-Week Exposure Cases		
K-400	500	188	53	8
Control	0	--	--	--

*Source: Ito et al., 1974

In the case of Kanechlor 400, all three of the dietary levels produced weight changes which do not seem to be normal; since control weights were not given for the Kanechlor groups this is more of an educated guess than a reported fact. It seems likely, however, that an increase of 23% in weight over the 40-week period is lower than would be expected. At the highest dose, 3/10 of the animals showed nodular hyperplasia and at 100 ppm 2/16 cases of nodular hyperplasia appeared. These findings were made at 40 weeks. This study was defective with respect to short duration and few animals used in the experiment.

While the investigators considered the nodular hyperplasia to represent preneoplastic lesions and to indicate the tumorigenicity of the Kanechlors, the experiment falls short of demonstrating tumorigenicity. The study is inconclusive because of the short duration, small starting numbers of animals in each group and still smaller numbers of animals at risk. However, the nodular hyperplasia which appeared as early as 40 weeks precludes adding this study to a negative finding.

The investigators found that their previous hypothesis that the effectiveness of the Kanechlor in producing tumors was a function of the level of chlorination of the PCBs in the mixture was not born out by this study.

The carcinogenicity of PCBs was investigated in a study sponsored by NCI (1978). Groups of 24 F344 rats of each sex were fed diets containing Aroclor 1254 at 25, 50 or 100 pm for 104-105 weeks.

Proliferative lesions in the liver were not found in controls but were found in the treated animals as shown in Table V-12.

TABLE V-12

Proliferative Lesions of the Liver in Fischer Rats Fed Aroclor 1254*

	Males		Females	
	Hepatocellular Carcinoma	Hyperplastic Nodules	Hepatocellular Carcinoma	Hyperplastic Nodules
Controls	0	0	0	0
25 ppm	0	5/24	0	6/24
50 ppm	1/24	8/24	1/24	9/22
100 ppm	3/24	12/24	2/24	17/24

*Source: NCI, 1978

The data indicate a dose-related nodular hyperplasia in both sexes and 3 cases of hepatocellular carcinoma among treated males. Although the incidence of hepatocellular carcinoma was not statistically significant it is important to consider the following points.

- In a preliminary study using 10 Sherman rats of each sex, Kimbrough (1972) did not produce hepatocellular carcinomas with doses of 100, 500 or 1000 ppm administered for a duration of less than a year. But in the other study (Kimbrough et al. 1975), which lasted 2 years, female Sherman rats had a 14% incidence of hepatocellular carcinoma as a result of treatment with 100 ppm Aroclor 1260. In the NCI (1978) study the incidence of hepatocellular carcinoma at 100 ppm Aroclor 1254 was given as 2/24 or 8%. (Actually, by 75 weeks on test in the 100 ppm group of males only 20 animals were alive. Since incidence is more precisely expressed as the number of animals with tumor/number of animals at risk, the figure is probably closer to 2/20 or 10%.) In either case, the incidence of 8-10%, or even 14%, as was found in the Kimbrough et al. (1975) study, would not be statistically significant with group sizes of 24 animals. The test was simply not sensitive enough to detect statistically significant differences at this level of potency. The lack of statistical significance in the NCI study has to be understood in terms of the highly insensitive nature of the assay used. A system which employed 24 animals per group would require a true difference in incidence between controls and treated groups of 35% in order to have a 90% chance of being significant at the p=.05 level. A 90% chance of finding a significant difference at the p=.05 level when the difference between controls and treated incidence is of the order of 9%, as it is in this study, would require 117 animals each for control and treated groups. It is therefore a question of biological significance, rather than statistical significance, in determining whether an 8-10% cancer yield represents a positive finding. Although this yield could not be used to demonstrate the carcinogenicity of the test agent, it is indeed consistent with the 14% yield obtained by Kimbrough et al. (1975) and lends support to that finding.

The NCI (1978) data were re-examined by Morgan et al. (1981) with respect to gastric adenocarcinomas which occurred in the rats in the NCI study. All 191 stomachs of rats from that study were available for further sectioning and examination. Sectioning at the pyloric junction revealed "lesions" upon gross examination in 3/47, 5/48, 8/48 and 17/48 of rats fed diets containing 25, 50 or 100 mg/kg Aroclor 1254, respectively. There did not seem to be sex-related differences in the incidence of these lesions. Histologically, adenocarcinomas were found in 1, 3, and 2 of the stomachs identified as having lesions in the 0, 25, 50 or 100 mg/kg Aroclor 1254 diet groups, respectively. Remaining lesions were identified as "metaplasia", the significance of which was not stated. Determining the occurrence of gastric adenocarcinoma in control rats to be <2% (0/47), the authors calculated the likelihood of six carcinomas occurring in stomachs of 144 treated animals to be <5% if occurrence was random. Morgan et al. (1981) concluded that Aroclor 1254 may produce adenocarcinomas in the stomachs of rats.

Mouse Studies -- Kanechlors 500, 400 and 300 were administered to male dd mice for 32 weeks at dietary levels of 500, 250 and 100 ppm. The nine different experimental groups had 12 animals/group and the controls had 6 animals/group. All of the animals survived the entire 32 weeks of the test and there was no apparent weight difference between control and treated animals although liver weight as a percentage of body weight was elevated in all treated groups.

The experiment was first reported in a brief note by Nagasaki (1972) and the results were given as grossly observable nodular hyperplasia in 7/12 (58.3%) of the animals fed 500 ppm of the Kanechlor 500. Microscopic examination revealed 5/12 (41.7%) of these animals had hepatomas. There were no tumors in the control animals or other test groups. In a more detailed report of this experiment Ito et al. (1973) gave the yield of liver tumors as 7/12 (58.3%) liver nodules and 5/12 (41.7%) hepatocellular carcinoma. The nomenclature of Ito (1973) is preferred because of the clearly preliminary nature of the Nagasaki (1972) note.

The question arises as to whether this study can be used for risk assessment. The composition of the Kanechlor 500 was given as 55% penta-CB, 26.5% TCB, 12.6% HCB and 5.0% bi-CB. This crude analysis does not entirely express the heterogeneous nature of the mixture of any of the commercial preparations. The 55% penta-CB will consist of a large number of isomers as will each of the other congener subgroups. The biological significance of this heterogeneity is that each of the isomers has its own particular toxicokinetic, metabolic and enzyme induction profile which is as much a function of the position of its chlorine substituents and the total number of chlorine substituents it contains. The extent to which any particular isomer contributes to or antagonizes the carcinogenic process is not known. The resultant carcinogenicity observed when the mixture is administered may be the work of one individual isomer, present in trace quantity or to the presence of another present to the extent of 25% of the composition of the mixture. What is not known in any of these studies is the actual dose of the carcinogen given the particular tumor yield. The tumor yield for any particular test is, therefore, a result of feeding that particular lot of a mixture which is different from any other lot of the same name.

The study demonstrates that that lot of Kanechlor 500 is capable of producing hepatocellular carcinoma in male dd mice when given at 500 ppm in the diet. The carcinogenic potential of the Kanechlor 500 is qualitatively demonstrated but quantitative application of the information obtained requires some considerations not immediately obvious from a statement of exposure level and tumor yield.

Kimbrough and Linder (1974) fed Aroclor 1254 to 2 groups of male Balb/cJ mice. There were 50 mice per treatment group and two groups of control mice (50 animals each). The treated groups each received 300 ppm of Aroclor 1254 in their diet; one group received the PCB containing diet for 11 months and the second group received the PCB containing diet for 6 months followed by a 5 month recovery period in which they received a normal diet.

There were a large number of deaths in the experiment which were a result of animal housing conditions and not due to toxicity.

At the end of 11 months the surviving animals were sacrificed. Only those animals that showed gross abnormalities were examined microscopically.

The livers of animals treated with the Aroclor were enlarged. Control animals had liver weights which were, on the average, 6% of their body weight. Treated animals had livers which were 7.5% of their body weight for the group exposed for 6 months and 25.5% of their body weight for those exposed for 11 months. Livers of treated animals showed multiple abnormalities including abnormal porphyrin metabolism as indicated by UV fluorescence. Adenofibrosis was also found among treated mice. The latter lesion is considered by some investigators to represent a premalignant lesion.

At the end of 11 months the surviving animals were sacrificed and the incidence of tumors of the liver was: 0/34 and 0/24 for the two control groups and 9/22 (40.1%) (10 hepatomas in 9 animals) in the 11 month exposure group; 1/24 (4%) of the animals in the 6 month exposure group had hepatomas. It should be noted that the Balb/cJ mouse has a low spontaneous incidence of hepatoma strain.

The experiment provides positive evidence that Aroclor 1254 is capable of producing a 40% incidence of hepatoma in male Balb/cJ mice at a dosage level of 50 mg/kg/day given for 11 months. The study provides confirmatory evidence of the carcinogenicity of commercial PCB mixtures in mice. In this case 300 ppm Aroclor 1254 produced 40% incidence of hepatoma in male Balb/cJ mice in 44 weeks and in the Ito et al. study (1973) Kanechlor 500 at 500 ppm in the diet for 32 weeks produced a 40% incidence of hepatocellular carcinoma in male dd mice.

The mouse studies, therefore, give strong evidence of PCB carcinogenicity. Table V-13 summarizes the findings on the PCB feeding studies with respect to liver tumorigenicity and carcinogenicity.

TABLE V-13

Effects on Liver Tumorigenesis and Carcinogenesis: Feeding Studies Using Various Commercial PCB Preparations

Species/ Strain/Sex	Agent	Duration	Exposure (ppm)	No. Animals	Results and Comment	Reference
Rats/Sherman (F)	Aroclor 1260	90 weeks	100	200/group	Hepatocellular carcinoma 14% (26/184) compared with 0.58% (1/173) controls, 78% neoplastic nodules compared with 0 in controls, no other treatment-related toxicity, study well done but limited to single dose and to females: shows carcinogenicity in female rats of Aroclor 1260.	Kimbrough et al., 1975
Rats/Fischer 344 (M&F)	Aroclor 1254	104-105 weeks	100 50	24/group	In males at 100 ppm, 8-10% hepatocellular carcinoma. Not statistically significant but study design would have required 35% or greater incidence for statistically significance. Results consistent with Kimbrough et al. (1975) and supports effect in males.	NCI, 1978
Rats/Donryu (M&F)	Kanechlor 400	22-77 weeks	38-462	10/group	Excessive toxicity, MTD exceeded, adenomatous nodules in females with total dose of 1200 mg or more; test too short, too few animals at risk, excessive toxicity. Test inadequate.	Kimura and Baba, 1973
Rats/Wistar (M)	Kanechlors 500 400 300	28-52 weeks	1000 500 100	22-25?/group	Nodular hyperplasia with Kanechlor 500 and Kanechlor 300, not statistically significant, duration of study too short, excessive toxicity. Test inadequate.	Ito, 1974
Mice/dd (M)	Kanechlors 500 400 300	32 weeks	500 250 100	12/group	Hepatocellular carcinoma with Kanechlor 500 at 500 ppm 41.7% (5/12) and liver nodules in 58.3% (7/12), demonstrates carcinogenicity in male mice.	Ito, 1973
Mice/Balb/cJ (M)	Aroclor 1254	1) 44 weeks 2) 24 weeks + 5 months recovery	300	50/group	Hepatoma 40.1% (9/22)*, none in controls in 44-week group	Kimbrough and Linder, 1974

*Animal deaths not due to toxicity but to housing. Demonstrates tumorigenicity of Aroclor 1254 in mice.

Conclusions Based on Feeding Studies -- Hepatocellular carcinomas were produced in female Sherman rats on a diet containing 100 ppm or Aroclor 1260 for 90 weeks and from feeding male dd mice Kanechlor 500 at 550 ppm for 32 weeks. The carcinogenicity of two different PCB preparation is, therefore, demonstrated in two species. Further support for the finding comes from the study on male and female F344 rats where some cases of hepatocellular carcinoma occurred in male rats given Aroclor 1254 at 100 ppm in the diet for 104 weeks. Although the results were not statistically significant, the sensitivity of this test was such that it would have been necessary to have an incidence of 35% over background in order to show statistical significance. The result was consistent with the study on Aroclor 1260 where the 14% incidence in a population of 184 rats was statistically significant. The study in F344 rats gave 8-10% hepatocellular carcinoma. In addition, another study in a strain of mice with a low spontaneous incidence of liver tumors, Balb/cJ, gave a 40.1% incidence of hepatoma with Aroclor 1254 given at a level of 300 ppm for 44 weeks.

The carcinogenicity of Aroclor 1260 and of Kanechlor 500 appears to have been demonstrated. The tumorigenicity of Aroclor 1254 has been shown and its carcinogenicity is suggested.

The qualitative picture seems to be clear with respect to several different commercial preparations of PCBs. The quantitative assessments which one would like to be able to make from the test data, however, present problems which severely limit the confidence one could place in such assessments. These difficulties in quantitative assessment spring from the nature of the commercial preparations used in the carcinogenicity tests. A discussion of these preparations and the implications of the facts follow.

__Difficulties and Considerations in Evaluating the Carcinogenic Potency of the PCB Preparations Tested__ -- The manufacturing process for commerical PCB products, such as the Aroclors, yields products composed of a mixture of 20-60 different polychlorinated biphenyl molecules. Individual lots of Aroclors of the same average chlorine content, and hence considered as the same commercial product, differ greatly in both their components and amounts of each component. The extent to this variable composition can be seen from the analyses of 3 different Aroclor 1260 preparations carried out in different laboratories. One preparation contained 26 PCBs, another 48 different PCBs and the third was only partially analyzed. The number of isomers found in the preparations for each level of chlorination is shown in Table V-14.

Not only are the number of isomers different, there is relatively little correspondence in the different preparations of the particular isomers present or the amount of any given isomer. Table V-15 shows data on the composition of only the hexachloro- isomers present in the 3 Aroclor 1260 preparations shown in Table V-14.

The degree of variability found in the hexachloro- isomer composition is found at other chlorination levels as well. Further, the variability is not confined to Aroclor 1260, but this product was used as an example because the best animal carcinogenicity data available (Kimbrough et al., 1975) was a study in which Aroclor 1260 was used.

In addition to the variability in polychlorinated biphenyls in the commercial PCB preparations, there are also a number of impurities in the products. Among the impurities are polychlorinated dibenzofurans (PCDFs) which are highly toxic and are under test for carcinogenicity.

TABLE V-14

Number of Isomers at Each Chlorinated Level for Three Different Aroclor 1260 Preparations*

Chlorination Level	Albro, 1976	Sissons and Welti, 1971	Tas and Vos, 1971
Dichloro-	0	7	NR
Trichloro-	0	7	NR
Tetrachloro-	2	3	NR
Pentachloro-	8	7	NR
Hexachloro-	5	8	4
Heptachloro-	6	5	3
Octachloro-	3	7	NR
Nonochloro-	2	3	NR
Decachloro-	0	1	NR
Total	26	48	NR

*Source: Adapted from NIOSH, 1977

TABLE V-15

Comparisons of Hexachlorobiphenyl Isomers of Aroclor 1260 Showing the Qualitative and Quantitative Variation in Three Different Aroclor Preparations*

Hexachlorobiphenyl Isomers Found in:		
3/3 Preparations	2/3 Preparations	1/3 Preparations
2,2',4,4',5,5' A - <4 B - major peak C - minor peak	2,3,3',4,4',5 A - <9 B - minor peak	2,2',3,4',5,5' A - <16
2,2',3,3',6,6' A - 4 B - major peak C - minor peak	2,2',3,4,4',5' B - major peak C - minor peak	2,2',3,4',5,6 A - <15
	2,2',3',4,5,6' B - major peak C - minor peak	2,3,3',4,4',6 B - minor peak
		2,2',3,3',4,4' B - major peak
		2,2',3,3',4,6' B - major peak

*Source: Adapted from NIOSH, 1977

A = Albro, 1976; B = Sissons and Welti, 1971; C = Tas and Vos, 1971

Numerical values in %

Several points concerning the interpretation of carcinogenicity data, and risk assessments based on the data, hinge upon the recognition of the qualitative and quantitative variability among commercial preparations.

1) Data on purified isomers shows that the metabolic and pharmacokinetic behavior of the different component molecules varies not only with the degree of chlorination, but also with the position of the chlorine atoms. Examples of how these findings bear on quantitative evaluation of the carcinogenic potency of PCBs follow.

2) Metabolism of purified isomers has been extensively studied. The hydroxylated metabolites of some 18 different individual PCBs were analyzed. It appears that predictable patterns of hydroxylation occur that indicate that the major pathway for most of the molecules involves direct hydroxylation. At least three different molecules involves direct hydroxylation. At least three different molecules among those studied produced products that indicated utilization of an alternate pathway in part. These three compounds were 4,4'-dichloro-, 2,2' 5,5'-tetrachloro-, and 2,2', 4,4', 5,5'-hexachlorobiphenyl. The alternate pathway for these three compounds involves the formation of arene oxide intermediates. Such intermediates would be expected to be carcinogens and mutagens based on studies on well known carcinogens. If the entire carcinogenicity of the commercial preparations was due to these components the potency of the preparations could be calculated on the amount of these isomers present, the percentage of parent compound that utilized this alternate pathway and the pharmacokinetics of the intermediates

formed. Table V-16 shows the results of analyses of 3 different Aroclor 1260, and 3 different Aroclor 1254 preparations for the presence of these arene oxide forming compounds.

The Aroclor 1260 data and Aroclor 1254 data were shown because these are commercial preparations which showed animal carcinogenicity. There is clearly little uniformity in the amounts of these compounds present, and also somewhat less of these 3 compounds altogether in the 3 Aroclor 1260 preparations analyzed than in the Aroclor 1254 preparations. Since only a small fraction of the parent compound utilizes the arene oxide pathway, if the entire carcinogenicity of the commercial mixture was attributable to these components, the potency might be at least a thousand fold greater than were the carcinogenicity due to all of the components in the mixture. (If the active agent comprised 1% of the mixture, and as much of 10% of this active component formed arene oxides, the active material would be .001 of the composition). One test in the Ames system with 2,2',5,5'-tetrachlorobiphenyl was negative, and 4 separate studies on Aroclor 1254 was negative in the micronucleus test, in the Dominant Lethal test, Sister Chromatid Exchange test, and in several tests for chromosomal abnormality. If the carcinogenicity observed with the Aroclors is due to the initiating activity fo epoxides which may be formed as metabolic intermediates, then the activity in the preparations is too low to be dected _in vitro_, or requires other _in vivo_ conditions to be expressed.

TABLE V-16

Analyses of Three Different Aroclor 1260 and Three Different Aroclor 1254 Preparations for the Presence of PCB Isomers Known to Utilize, to Some Degree, A Metabolic Pathway which Forms Arene Oxide Intermediates

Aroclor	4,4'-Dichloro-	2,2',5,5'-Tetrachloro-	2,2',4,4',5,5'-Hexachloro-
1260 (Albro)	absent	absent	<4%
1260 (Sissons)	minor peak	minor peak	major peak
1260 (Tas)	*	*	minor peak
1254 (Albro)	absent	8%	4%
1254 (Sissons)	minor peak	major peak	major peak
1254 (Willis)	absent	minor peak	minor peak

*Probably not analyzed

The metabolic data, along with the information on chemical analyses, and the *in vitro* tests all suggest that if these components do act as initiators their role in the carcinogenicity may be contributory but is unlikely to be the sole mechanism involved. It is, therefore, unsatisfactory to calculate the potency on the basis of exposure to "possible" active components, or to calculate the potency on the basis of exposure to any of the other components, some of which are scarcely metabolized at all.

3) The analyses of different Aroclor 1260 lots and different Aroclor 1254 lots indicates that a large percentage of Aroclor 1260 would consist of penta-, hexa- and heptachloro- isomers. For example, the bulk of a particular Aroclor 1260 was composed of 2 each of penta-, hexa- and heptachloro- isomers; another Aroclor 1260 lot had 11 major peaks consisting of 2 penta-, 6 hexa- and 3 heptachloro- isomers. An Aroclor 1254 had over 20% of a single penta- isomer and 14% or more of 3 other penta- isomers and 12% of a hexa- isomer. The pharmacokinetics of penta-, hexa- and heptachloro- compounds is, therefore, pertinent to the assessment of total body burden of PCBs of treated animals. The data show that a particular hexachloro- isomer (2,2',4,4',5,5'-) is largely distributed in skin and adipose tissue. As long as 42 days after i.v. administration of this isomer there was still 85% of the administered dose in adipose tissue, less than 1% of which was metabolite. In other words, a hexachlorobiphenyl isomer, known to be metabolized, is still largely retained as parent compound in tissue over a month after administration. The several hexachloro- isomers found in the commercial preparations can be expected to be retained at least as long as this particular hexachloro- isomer.

In contrast, a tested pentachloro isomer was retained in adipose tissue to only a small degree (2.7% at 42 days following administration).

These observations provoke the question of what common effect Aroclor 1260 and Aroclor 1254 can have on the liver since the bulk of the Aroclor 1254 is pentachloro, and would be expected to be relatively rapidly metabolized and excreted, while the major components of Aroclor 1260 are hexa- and heptachloro- isomers which would be retained for long periods in storage depots such as sking and adipose tissue. Furthermore, it is not clear to what extent stored hexa- and hepta compounds constitute effective "removal" of the agent or conversely constitute a "carcinogen pool" capable of being mobilized and adding to the liver burden in a continuous feeding study.

In either case the administered dose, whether calculated on an average daily dose, or as a total dose, does not represent a realistic measure of exposure or body burden of PCB. A comparison of an Aroclor 1260 and an Aroclor 1254, given in the same daily dose would result in completely different internal tissue specific exposure levels particularly in the liver which is the target organ for the carcinogenic effect.

This consideration demonstrates the inappropriateness of a potency calculation which considers the exposure to be a simple function of the amount of PCB administered in the diet.

4) One of the most striking findings concerning the variability of the components of the commercial products is the differing enzyme inducing capacities of particular isomers even at the same level of chlorination. The enzymes induced range from those which are involved in metabolism of PCBs themselves to others which have been implicated as activators and inactivators of other procarcinogens or carcinogens respectively. The level of enzyme induction reflects the concentration of the inducer. The variable concentration of the many congeners and isomers in any lot of an Aroclor is, as would be anticipated, assurance that the pattern of induction and level of induction of liver enzymes will be a unique expression for that particular lot tested. It will represent the total of many different activities, some of which will be capable of reducing the carcinogenic effect and some of which will increase the carcinogenic effects.

5) These examples show why the test data on the carcinogenicity of PCBs generated by use of commercial preparations such as the Aroclors and Kanechlors can provide only a net effect picture of the many and varied effects of the individual components in the preparations. The carcinogenicity which is manifest reflects the resultant of vectors which represent partial additive, synergistic and/or antagonistic effects of numerous individual components. We have shown that the experimental data indicate that it is inappropriate to use data from commercial preparations composed of 20-50 different compounds with differing metabolic, pharmacologic and pharmacokinetic properties to calculate potency and exposure in a scientifically valid way. Since

potency and exposure are basic parameters used in risk estimation, it follows that such estimation should be undertaken only after the active agent or agents have been identified and studied.

6) It can be said, however, that it is very likely that the potency of any commercial PCB preparation is considerably higher than any figure obtained by utilizing the dietary level of exposure as a basis for calculation.

Other Studies -- A report published by DiGiovanni et al. (1978) in which 100 μg of Aroclor 1254 was applied to the skin of CD-1 mice prior to application of DMBA followed by repeated applications of a phorbol ester promoter, stated that that data showed weak initiator activity.

The tumor promoting activity of Aroclor 1254 was investigated in a study by Berry et al. (1978). Groups of 30 female CD-1 mice were used. The animals were initiated with 0.2 m DMBA; one week later a positive control received 2 μg TPA and an experimental group received 100 μg Aroclor 1254. The promoter and the Aroclor applications were made twice weekly for 30 weeks. DMBA promotion resulted in 92% of the animals with papillomas while none of the Aroclor 1254 treated animals developed tumors. It was concluded that at the dose of Aroclor 1254 used in the study the preparation was not a skin tumor promoter in this system. General toxicity precluded use of larger doses in the experiment.

In a test carried out by Ito et al. (1973) Kanechlor 500 was evaluated in combination with 2, 3, 8 or benzenehexachloride (BHC). The table showing the results follows (Table V-17).

TABLE V-17

Action of Kanechlor 500 and α-, β- or γ-Benzene Hexachloride*

Test Compound(s) (ppm)	Nodular Hyperplasia (%)	Hepatocellular Carcinoma (%)
α-BHC (250)	23/30 (76.7)	8/30 (26.7)
α-BHC (250) + PCB (250)	21/30 (80.8)	15/30 (57.6)
α-BHC (100)	0/26	0/26
α-BHC (100) + PCB (250)	8/25 (32.0)	1/25 (4.0)
α-BHC (50)	0/28	0/26
α-BHC (50) + PCB (250)	9/30 (30.0)	2/30 (6.7)
β-BHC (250)	0/26	0/26
β-BHC (250) + PCB (250)	16/29 (55.2)	6/29 (20.7)
β-BHC (100)	0/26	0/26
β-BHC (100) + PCB (250)	5/30 (16.7)	1/30 (3.3)
β-BHC (50)	0	0
β-BHC (50) + PCB (250)	0	0
γ-BHC (250)	0	0
γ-BHC (250) + PCB (250)	0	0
γ-BHC (100)	0	0
γ-BHC (100) + PCB (250)	0	0
γ-BHC (50)	0	0
γ-BHC (50) + PCB (250)	0	0
PCB (250)	0/20	0/20
Controls	0/20	0/20

*Source: Ito et al., 1973.

These data show that when male dd mice were given Kanechlor 500 (250 ppm) in their diet for 32 weeks with or without one of the BHC compounds, at 250 ppm, they responded by yielding a higher incidence of hepatocellular carcinoma than when either the BHC or the Kanechlor were given alone. This was true for the BHC and for the BHC at the 250 ppm level.

Although the title of the paper indicates that the experiment is a test for "promotion", it is not since both of the compounds were given concurrently in the diet. Further, since both of these agents are capable of producing hepatocellular carcinoma it is critical that the total dose be adjusted when both agents are given at the same time. The total dose of the two agents combined was 500 ppm, not 250 ppm. Therefore, although synergy may have been operative, as the authors suggested, the results are as easily interpreted as complementary or additive in nature. Neither promoting or synergistic effects were demonstrated by the tests.

In the study of Makiura et al. (1974) Kanechlor 500, at 0.05% in the diet for 24 weeks did not produce detectable tumors for the 24 weeks of the experiment. When this regimen was modified to add 3'-methyl-4-dimethylaminoazobenzene (3'-MeDAB), N-2-fluorenylacetamide (2-FAA), or diethylnitrosamine (DEN) at carcinogenic levels the combined effect was to reduce the incidence of tumors to zero. In the Sprague-Dawley rats used the tumor incidence with these 3 agents was 65% (3'-MeDAB), 54% (2-FAA) and 92% (DEN) liver tumors. In paired combination 3'-Me-DAB and DEN gave 92% liver tumor incidence which was reduced to 7.7% when PCB was also given. When 2-FAA and DEN were given together the tumor yield was 82% and in the experiment with these 2 agents plus PCB the yield was zero. There is, however, a problem with the experiment which modifies what appears to be an obvious conclusion.

The combined agents produced extreme toxicity as shown by the weight records, shown in Table V-18.

The data show that while each agent alone produced some reduction in growth compared to controls, the combined effects were substantial. It is not possible to tell from this experiment whether the effects observed on tumor growth were due to a general systemic debilitation or whether the effects represent specific drug-related responses. The overall toxicity of the combined agents was not adjusted so that it would be no greater than the level of toxicity of each alone.

Nishizumi (1980) demonstrated the ability of placentally transferred PCBs to inhibit DEN-induced hepatomas. Groups of 10, 10-week-old female Wistar rats were treated with 200 or 40 mg/kg bw/day Kanechlor 500 (Ito, 1973) by gavage on days 5, 10 and 15 of gestation. A group of 10 olive oil treated rats served as vehicle controls. At 28 days of age, one F_1 offspring from each litter was killed for quantitation of liver PCBs. Remaining F_1 offspring were exposed to 50 mg/ℓ DEN in drinking water continuously for 5 weeks. At 16, 20 and 24 weeks after the start of exposure to DEN, 6-8 F_1 rats of each sex from each treatment group were killed, liver weight was recorded as percent of body weight and livers were sectioned and grossly examined for "tumors" $\geq$5 mm diameter.

TABLE V-18

Percent Weight Change in Kanechlor 500 Exposed Rats*

Weight Change %		Weight Change %		Weight Change %	
Control	+180	Control	+180	Control	+180
3-MeDAB	+150	PCB + 3-MeDAB	+107	PCB + 3-MeDAB + DEN	+65
DEN	+137	PCB + DEN5	+73	PCB + 2FAA + DEN	+52
2FAA	+116	PCB + 2FAA	+100		
PCB	+112				

*Source: Adapted from Makiura et al., 1974

MeDAB = 3'-methyl-4-dimethyl-aminoazobenzene; DEN = Diethylnitrosamine; 2FAA = N-2-fluorenylacetamide

Dams showed no signs of maternal toxicity during gestation or the following 6 months (Nishizumi, 1980). There was no evidence of fetal toxicity; average litter size was 6.2, 6.5 and 7.1 in the 400, 40 and 0 mg/kg groups, respectively. The 500 mg/kg groups experienced several losses of F_1 progeny (numbers not reported) and slight depression of rate of gain (body weight not given) before weaning.

The authors reported that almost all liver nodules >5 mm in diameter were histologically hepatocellular carcinomas and that some nodules >5 mm were neoplastic nodules (number not reported) (Nishizumi, 1980). Number of nodules $\geq$5 mm diameter was therefore chosen as the endpoint indicator of hepatocarcinogenic potency. The result of this study are summarized in Table V-19. Liver weight as percent of F_1 body weight did not differ statistically between treatment and control groups. Although liver weight and average number of liver tumors/rat were reported as separate figures for male and female F_1 progeny, it was unclear if statistical analysis compared treatment animals to control animals by sex or collectively. Significant ($p<0.05$) reduction in number of liver tumors after 20 weeks occurred in both sexes of rats in the high-dose group, and in the low-dose group males.

At the 24-week examination, significant ($p<0.05$) reduction in tumor incidence occurred only in high-dose group males (Table V-19). These data are presented in Table V-19. The authors reported liver PCB values in 28-day-old F_1 individuals of 360±30 ppm, 18±7 ppm, and <1 ppm for high-dose, low-dose and control groups, respectively. They, therefore, suggested that placental transfer of PCBs protected the treatment rats from DEN-induced tumors.

TABLE V-19

The Liver and Body Weight Ratio and the Number of Liver Tumors Produced in Offspring Rats Exposed to PCB through Their Dams and Treated with DEN after Weaning[a,b]

Group No.	Treatment		Sex of Offspring	Liver Weight as Percent Body Weight		Average Number of Liver Tumors (>^{a} mm)/Rat	
	Dam	Offspring		20 week	24 week	20 week	24 week
1	PCB 200/mg/kg 3 times	DEN 50 ppm 5 weeks	male female	5.3 ± 0.1 5.1 ± 0.3	6.4 ± 0.3 6.2 ± 0.2	1.0 ± p04^{c} (6,4,6) 0^{c} (0,0,8)	2.0 ± 0.7^{c} (14,5,7) 0.4 ± 0.3 (3,2,8)
2	PCB 40/mg/kg 3 times	DEN 50 ppm 5 weeks	male female	5.3 ± 0.2 5.5 ± 0.1	6.3 ± 0.5 5.8 ± 0.2	1.3 ± 0.4* (10,6,8) 0.6 ± 0.3 (5,4,8)	2.8 ± 0.7 (17,6,6) 0.7 ± 0.4 (5,3,7)
3	None	DEN 50 ppm 5 weeks	male female	6.0 ± 0.4 5.6 ± 0.3	7.5 ± 0.6 6.1 ± 0.4	3.0 ± 0.7 (21,6,7) 1.1 ± 0.4 (9,5,8)	4.6 ± 0.7 (37,8,8) 1.4 ± 0.5 (10,4,7)

[a] Source: Nishizumi, 1980

[b] The data are expressed as means ± SE. Numbers in parentheses are the total number of liver tumors/group, the number of rats bearing liver tumors, and the number of rats sacrificed, in that order.

[c] Significant at the 5% level as compared with group 3.

In a brief communication of a short-term study, Ito et al. (1978) demonstrated the ability of PCB (mixture not stated or characterized) and other organic compounds to enhance nodular liver hyperplasia induced by feeding 2-FAA, a known liver cancer inducer, to rats. Male Fischer rats weighing ~155 g were used. Two groups of control rats were fed diets containing 200 mg/kg 2-FAA for the 10-week trial; one group was partially hepatectomized in the third week of the study. There were two PCB treatment groups, both fed a diet containing 1000 mg/kg PCB for the entire 10-week experimental period, and one group partially hepatectomized in the third week of the study. PCB-fed rats had liver containing significantly ($p<0.05$) more hyperplastic nodules/10 cm^2 than control rats. Partial hepatectomy significantly ($p<0.001$) increased the incidence of nodular hyperplasia in the treatment groups. These authors investigated this two-stage model of liver tumorigenesis as a possible short-term in vivo screening test for hepatocarcinogens.

A study of the cocarcinogenic effect of PCB feeding on 3-methylcholanthrene (3-MC) induced cervical cancer was performed in mice (Uchiyama et al., 1974). Adult virgin female dd mice were fed control diet or diet containing 10 ppm for 15 weeks or 100 ppm for 8 weeks of Kanechlor 400. Cotton thread impregnated with 3-MC was inserted into the cervix and through anterior horn, and left in place for 4 weeks. PCB did not increase the incidence of cervical epithelial change induced by 3-MC, although effective numbers surviving the experiment were small (20 mice in each PCB-fed group). A simultaneous study using 10 ppm and 100 ppm DDT showed considerable tendency to induce cervical carcinoma, which may be considered a positive control. In this trial using small numbers and a short experimental period, the authors found that PCB fed at these levels did not effectively enhance 3-MC-induced cervical carcinoma.

Comments on Other Studies -- Under the limited conditions of one test promoter activity was not demonstrated for Aroclor 1254. Another test purported to measure promoter activity did not, in fact, measure this behavior.

Antagonistic activity with respect to tumors produced by other agents may be a property of Kanechlor 500. The ability of an agent which under one set of circumstances induces tumors, to antagonize tumor induction or growth, should not be surprising. It has been well demonstrated that the various congeners found in commercial PCB preparations each evoke distinct and different liver enzyme profiles. A particular molecule in the mixture may induce enzymes which activate procarcinogens while another molecule in the mixture induces an enzyme cluster which hastens the metabolism of other ultimate carcinogens. Indeed this heterogeneity explains why the final manifestations in any particular test with a commercial PCB mixture represent NET effect of multiple components present in varying concentration in the mixture. This point has been discussed earlier under the heading of difficulties in quantitative risk assessment.

The various experiments discussed under "other studies" do not provide the critical studies or data which explain or clarify the mechanisms of action of PCB preparations.

Mutagenicity. Mutagenicity studies of PCBs in *Salmonella typhimurium* are summarized in Table V-20. PCBs have not shown positive results by themselves, but evidence exists that metabolic activation may result in mutagenic metabolites. Evidence also exists that the less chlorine substituted varieties may be more mutagenic. Hepatic S-9 fractions from some strains of rats may be better able to activate PCB metabolism than S-9 fractions of other strains.

Schoeny et al. (1979) tested Aroclor 1254 for mutagenicity at eight different dosage levels from 0.5-500 mg/plate in strains TA1535, TA15397, TA98 and TA100 of *S. typhimurium*. Aroclor 1254 alone, activated by S-9 hepatic fractions from uninduced or Aroclor 1254-induced Sprague-Dawley rats, failed to manifest mutagenicity. Subsequently, this same laboratory, recognizing the heterogeneous nature of commercial Aroclor, conducted a more extensive and well-controlled study of the mutagenicity of four separate PCBs (Schoeny, 1981). Using materials of 99% purity, eight doses of 4-chlorobiphenyl and five doses each of 3,4,3',4'-tetra CB; 2,4,2',4'tetra CB and 2,4,6,2',4',6'-hexa CB were tested for mutagenicity in *S. typhimurium* (see Table V-20). Mutagenicity was not demonstrated with any of these PCBs with or without the addition of hepatic S-9 fractions. S-9 fractions from rats induced with Aroclor 1254, phenobarbital, 3-MC or corn oil (vehicle control) were used with protein content varied to assure optimum concentration. In another part of the same study, the authors likewise failed to demonstrate mutagenicity of dibenzofuran or various polychlorinated dibenzofurans, often considered common contaminants of PCBs.

TABLE V-20

Summary of Mutagenicity Assays of PCBs in *Salmonella typhimurium*

Type of Assay	Type of S-9 Activation	PCB Source	PCB Dose	Strains of *S. typhimurium* and Result					Reference
				TA1535	TA1539	TA98	TA100	TA1538	
Plate	none	Aroclor 1254	0.5-500 µℓ/plate	-	-	-	-	NA	Schoeny et al., 1979
Plate	noninduced	Aroclor 1254	0.5-500 µℓ/plate	-	-	-	-	NA	
Plate	Aroclor 1254 induced	Aroclor 1254	0.5-500 µℓ/plate	-	-	-	-	NA	
Plate	none	4-chlorobiphenyl	1-200 µg/plate	NR[b]	NR	NR	NR		Schoeny, 1981
	PCB[a]			-	NA	NA	NA		
	PB[a]			NA	-	NA	NA		
	3MC[a]			NA	NA	NA	-		
	corn oil[a]			NA	NA	-	NA		
	PCB[c]	3,4,3',4'-tetra-chlorobiphenyl	5-200 µg/plate	NA	NA	NA	-		
	3MC[c]			NA	NA	-	NA		
	PCB[c]	2,4,2',4'-tetra-chlorobiphenyl	5-200 µg/plate	NA	NA	NA	-		
	3MC[c]			NA	NA	-	NA		
	PCB[c]	2,4,6,2',4',6'-hexachlorobiphenyl	5-200 µg/plate	NA	NA	NA	-		
	3MC[c]			NA	NA	-	NA		
Plate	rabbit liver microsomal preparations	4-chlorobiphenyl	50-200 µg/plate	NR	NR	NR	NR	++ +	Wyndham et al., 1976
		Aroclor 1221		NR	NR	NR	NR	-	
		2,2',5,5'-tetra-chlorobiphenyl		NR	NR	NR	NR	-	
		Aroclor 1254		NR	NR	NR	NR		

[a] At 0, 10, 50 and 100 µℓ/plate

[b] Text failed to state what strains were tested for mutagenicity without S-9; results were reported as negative.

[c] 50 µℓ/plate

NA = Not applicable; NR = Not reported

+ = Positive; - = Negative

One laboratory has attempted to relate degree of mutagenicity to degree of chlorine substitutions on the biphenyl moiety (Wyndham et al., 1976). Using levels of 10, 50, 100 or 200 μg/plate of the monochlorinated 4-chlorobiphenyl, Aroclor 1221 [average chlorine content 1.15 (2 Cl/molecule)], 2,2',5,5'-tetrachlorobiphenyl (4 Cl/molecule) or Aroclor 1254 (average 4.96 Cl/molecule), these authors demonstrated a pronounced mutagenicity (>2000 mutant colonies/plate) of 4-chlorobiphenyl compared with the more highly chlorinated PCBs. Although it was not clear from the text, it appeared that rabbit liver microsomal fractions were used for metabolic activation. No reason was given for this choice. Testing without metabolic activation resulted in mutagenicity only slightly greater than that evidenced by controls. Concurrent studies (see Enzyme Induction Section in Chapter VII) using ^{2}H and ^{3}H labeled 4-chlorobiphenyl elucidated that metabolism of 4-chlorobiphenyl proceeds by an arene oxide intermediate that binds to microsomal RNA and protein. These authors interpreted the data to suggest that the metabolism of 4-chlorobyphenyl occurs at a more rapid rate than the more highly chlorinated species; therefore, greater mutagenicity was observed.

Heddle and Bruce (1977) examined the mutagenicity of Aroclor 1254 in a group of 61 potential mutagens and compared mutagenicity with the production of cytogenetic effects in mice. The purpose of this study was to demonstrate a correlation between mutagenicity in S. typhimurium and cytogenetic abnormalities manifested as micronuclei and production of abnormal sperm. Details of both assays, including dosage levels and detailed results, were conspicuously lacking. These authors found Aroclor 1254 to be nonmutagenic in the S. typhimurium bioassay and negative in both cytogenetic evaluations. S. typhimurium strains TA1535, TA1537, TA98 and TA100 both with and without S-9 activation were tested.

One study examined dominant lethality in rats attributable to PCBs (Green et al., 1975b). Mature, random-bred Osborne-Mendel male rats were given Aroclor 1242 by gavage in single doses of 625, 1250 or 2500 mg/kg bw. In a second study, Aroclor 1254 was given by gavage at levels of 75, 150 or 300 mg/kg/day for 5 consecutive days. In a third study, male rats were given five daily doses of 125 or 250 mg/kg/day Aroclor 1242 or Aroclor 1254 at dosage levels of 75 or 150 mg/kg/day. Treatment groups consisted of 10 animals each; negative controls were treated by gavage with corn oil alone and positive controls were treated with a known mutagen, triethylenemelamine (TEM). Male rats were mated for 10-11 weeks and females were killed and examined for implantations and pregnancies at mid-pregnancy. Significant fetal loss occurred only in TEM-treated positive controls. Similarly, a feeding trial with Aroclor 1254 at levels of 25 or 100 mg/kg diet for 70 days produced no significant fetal loss. Positive controls were not used.

Nilsson and Ramel (1974) reported briefly on a study to determine clastogenic effects of PCBs in Drosophila melanogaster carrying sex-linked color markers. Clophen 30 (30% chlorine) or Clophen 50 (50% chlorine) was incorporated into the substrate at 250 or 200 mg/ℓ, respectively. Treatment was given during the entire larval period or during 1 week to adult flies. These levels were chosen on the basis of the results of preliminary trials using levels of 62.5, 125, 250 or 500 mg/ℓ substrate of Clophen 30 or levels of 25, 50, 100 or 200 mg/ℓ substrate of Clophen 50 to determine the toxic effects of these PCB products on larval development. The authors determined that the levels of Clophen products chosen for the genetic studies might slightly retard larval development but should not cause mortality or mask genetic effects. Nondisjunction giving rise to XXY females and XO males and chromosomal loss resulting in XO males were chosen as endpoint indicators of chromosomal aberration.

Individuals with chromosonal aberrations were immediately apparent because of the phenotypic expression of these sex-linked markers (Nilsson and Ramel, 1974). The Clophen 30-treated group contained 15/8403 (0.18%) XXY females and 52/7818 (0.67%) XO males. The Clophen-30 control groups contained 8/7096 (0.11%) XXY females and 53/6871 (0.77%) XO males. The Clophen 50-treated group contained 6/14316 (0.04%) XXY females and 76/12982 (0.59%) XO males. The Clophen-50 control group contained 19/15614 (0.12%) XXY females and 106/14499 (0.73%) XO males. Differences between the Clophen 30 group and its controls were not significant. The incidence of XO males in the Clophen 50-treated group was significantly ($p<0.01$) greater than its controls. A larger Clophen 50 trial was therefore performed using 20 mg/ℓ substrate larval treatment of parental males. Examination of 12 different broods of offspring revealed the presence of 11/37738 (0.03%) XXY females and 128/33085 (0.39%) XO males in the treatment groups and 23/68644 (0.03%) XXY females and 202/59064 (0.34%) XO males in the control groups. These authors concluded that the products tested did not indicate clastogenic effects but pointed out that these products differed from commercially available PCB products in that the impurities ordinarily present in commercial products were not present here.

A cytogenic study of Aroclor in rats was performed by Green et al. (1975a). Aroclor 1242 was given orally in single doses of 1250, 2500 or 5000 mg/kg bw or at multiple doses of 500 mg/kg/day for 4 days to groups of eight male, random-bred Osborne-Mendel rats. Acute debility, the result of cumulative toxicity, precluded dosing the last group for the intended 5 consecutive days. Aroclor 1254 was orally administered at 75, 150 or 300 mg/kg/days for 5 consecutive days to groups of eight rats. Corn oil-treated controls were used with each Aroclor tested. Rats were killed 24 hours after the last oral dose; femur bone marrow was aspirated from both Aroclor 1242- and Aroclor 1254-treated rats. Spermatogonia were processed only from Aroclor 1242-fed rats. Since Aroclor 1242 was the more toxic product and did not result in evidence of having caused cytogenetic damage in spermatogonia, these authors did not believe it necessary to examine spermatogonia from Aroclor 1254-exposed rats.

Weight loss occurred in rats treated with 1250 mg Aroclor 1242 (Green et al., 1975a). Significant (4/8, 3/8) mortality occurred in the two higher-dosed groups. Weight loss and mortality (1/8) occurred in rats exposed to 300 mg Aroclor 1254. Percentages of cells with chromosomal abnormalities and cells in mitosis were 0.5, 4.0%; 0.0, 3.2%; 0.3, 4.1%; 0.0, 4.2% and 1.2, 4.1% in bone marrow from rats exposed to 0, 500, 1250, 2500 or 5000 mg Aroclor 1242, respectively. These results were not significant. Percentages of spermatogonia with chromosomal abnormalities and in mitosis were 0.4, 1.5%; 1.0, 0.7%; 0.3, 1.3%; 0.5, 1.8% and 0.8, 0.9% from testes of rats exposed to 0, 500, 1250, 2500 or 5000 mg Aroclor 1242, respectively. Percentages of cells with chromosomal abnormalities and cells in mitosis in bone marrow were 0.5, 4.1%; 0.5, 4.0%; 0.3, 2.9% and 0.6, 3.1% in 0, 75, 150 and 300 mg/kg/day Aroclor 1254-treated groups, respectively. Again, numbers of observed chromsomal aberrations were not significant. The number of bone marrow cells observed in mitosis appeared to be depressed ($p<0.05$) in the Aroclor 1254 groups at the mid- (150 mg/kg/day) and high- (300 mg/kg/day) treatment levels. Mitosis in bone marrow cells of Aroclor 1242-treated groups did not appear to be depressed, but spermatogonial mitosis did appear to be depressed in the 500 mg/kg/day for 4 days ($p<0.01$)and the 5000 mg/kg ($p<0.05$) groups. The authors concluded that Aroclor 1242 and 1254 possessed "no mutagenic potential as assessed by cytogenetic analysis of rat bone marrow and spermatogonia." They felt that the small degree of mitotic inhibition observed was probably evidence for PCBs or metabolic products thereof actually entering the tissues studied, rather than a general toxic manifestation of PCBs on the organism as a whole.

A later cytogenetic study of bone marrow and spermatogonia in male Holtzman rats (Garthoff et al., 1977) confirmed the negative finds of the previously cited study. Aroclor 1254 was fed for 34-37 days at levels of 5, 50 or 500 mg/kg diet/day. Examination of cells from 9, 7, 7 and 6 rats, respectively, from the above groups revealed 0.0%, 3.03%; 0.3%, 0.0%; 0.0%, 4.4% and 1.0%, 0.0% chromosomal abnormalities in bone marrow cells and spermatogonia, respectively. Results were not dose-related and were statistically insignificant.

Likewise, Dikshith et al. (1975), in a more comprehensive study of the effects of orally administered Aroclor 1254 on rat testis, found no significant evidence of chromosomal aberration caused by PCBs. Aroclor 1254 was administered by gavage to 18 mature, male Sprague-Dawley rats at the level of 50 mg/kg bw daily for 7 consecutive days. A vehicle control group of 18 rats was similarly dosed with corn oil. Three rats from each group were killed and examined on days 1, 7, 15 and 30. Since the focus of this investigation was to study effects of PCBs on histology and histochemistry of rat testis, details of the protocol to evaluate chromosomal aberrations were not given. The authors simply stated that configurations of chromosomes appeared similar in both the control and treatment groups. A few chromosomal abnormalities were observed but appeared to be sporadic.

More recently, Watanabe and Sugahara (1981) investigated the mutagenicity of PCBs alone and in combination with dimethylnitrosamine (DMN) or N-methyl-N'nitro-N'nitrosoguanidine (MNNG). Kanechlor 500 was dissolved in corn oil or 95% ethanol and administered at the rate of 100 mg/kg bw/day for 6 days to 5-week-old male ddY mice weighing 25-30 g. The corn oil solutions were given by gavage and the ethanol solutions were given by subcutaneous injections. DMN (25 mg/kg bw) or MNNG (100 mg/kg bw)) in aqueous solutions were administered 24 hours after the last dose of PCB. The number of cells containing micronuclei observed in polychromatic erythrocytes (PCE)/1000 cells counted from femural bone marrow taken 30 hours after DMN or MNNG was given was considered the endpoint of mutagenicity. The DMN treatment groups and numbers of mice/group were as follows: corn oil control, 3; PCB in corn oil, 9; DMN alone, 5; corn oil + DMN, 4; PCB in corn oil + DMN, 9. A total of 4000 cells counted/mouse yielded the following results expressed as number of micronucleated cells/1000 PCE counted in the above-described five groups, respectively: 0.9, 1.6 ($p<0.05$), 7.7, 3.9 ($p<0.01$) and 4.7. The authors concluded that these results indicated that corn oil interfered with the expected mutagenicity of DMN. They therefore restructured the experiment so that PCBs were dissolved in ethanol and administered by subcutaneous injections to groups of mice as follows: ethanol alone, 5 mice; PCB in ethanol, 5 mice; DMN alone, 6 mice; ethanol + DMN, 5 mice; PCB in ethanol + DMN, 5 mice. Counting the same numbers of PCE as previously described revealed the following numbers of micronucleated cells/1000 counted: 0.9, 1.5, 8.0, 8.6 and 7.4. These results of PCB treatment did not differ significantly from negative or positive controls and the authors concluded that Kanechlor 500 had "almost no effect" and no synergistic or addition effect with DMN. PCB in ethanolic solution injected subcutaneously was combined with MNNG as follows: ethanol alone, 2 mice; PCB in ethanol, 3 mice; MNNG alone and bone marrow examined at 24 hours, 3 mice; MNNG alone and bone marrow examined at 30 hours, 3 mice; ethanol + MNNG and bone marrow examined at 24 hours, 3 mice; ethanol + MNNG and bone marrow examined at 30 hours, 3 mice; PCB in ethanol + MNNG and bone marrow examined at 24 hours, 5 mice; and PCB in ethanol + MNNG and bone marrow examined at 30 hours, 4 mice. Counting 4000 PCE/mouse and expressing micronucleated cells as number/1000 PCE yielded the following results in the above-detailed groups: 0.6, 0.9, 1.6, 2.0, 1.4, 1.7, 1.3 and 1.7, respectively. PCBs alone or in combination with MNNG failed to increase numbers of micronucleated PCE. Although data were not presented on numbers of micronucleated PCE/1000 cells for individual mice, the authors stated that considerable variation existed. Each cell preparation was examined by two individuals and, in some cases, significant variations between examiners existed.

Cytogenetic examinations were performed on 3- to 6-day-old ring dove (Streptopchia risoria) embryos from doves fed Aroclor 1254 for an extended length of time at the level of 10 ppm of diet (Peakall et al., 1972). Negative controls and positive controls (x-ray treated) were used. Chromosomal aberrations were recorded for the largest eight chromosome pairs occurring in metaphasic allantois and limb bud cells. An average of 365 cells/embryo were examined. Cells manifesting chromosomal aberrations were 0.8, 1.8 and 18.0% for negative control, Aroclor 1254- and x-ray-treated groups, respectively. The authors considered these results indicative of a possible clastogenic action of PCBs in dove embryos.

Teratogenicity and Reproductive Toxicity.

Effects on Conceptus of Rodents and Rabbits -- Several studies have been reported investigating the teratogenetic and fetotoxic effect of various mixtures of PCBs. Little evidence of teratogenicity was found; most reportshad to do with the fetotoxic potential of PCBs and their ability to interfere with expected levels of fertility in many mammalian species. The more pertinent studies are summarized in Table V-21.

One of the first reports (Villeneuve et al., 1971) examined fetal toxicity of Aroclor 1254 in the rabbit and rat. Doses of 0, 6.25, 12.5, 25, 50 or 100 mg/kg/day of Aroclor 1254 in corn oil administered on days 6-15 of pregnancy to groups of 10 Wistar rats failed to produce maternal toxicity (evaluated as body weight gain, fertility and litter size) or fetal toxicity (evaluated as number of litters, average litter size, resorption sites/litter, average litter weight, skeletal or visceral abnormalities). The 100 mg/kg treatment resulted in litters with average pup weights significantly lower than controls. However, total litter weight (average weight times number of fetuses) did not differ from controls.

TABLE V-21

Summary: Teratogenetic, Fetotoxic and Reproductive Effects of Orally Administered PCBs

Species/ Strain	Source of PCBs	Dosage Level and Duration of Treatment	Maternal Response	Progeny Response	Reference
Rabbit	Aroclor 1254	0, 1.0, 10.0, 12.5, 25.0, 50 mg/kg/day on days 1-28 of pregnancy; 25 mg/kg/day on days 7-28 of pregnancy	Dose ≥25 mg/kg: maternal death, weight loss; ≥10.0 mg/kg: hepatomegaly; 25 mg/kg days 7-28: reduced rate of gain.	≥12.5 mg/kg/day: fetal death, resorptions, abortions.	Villeneuve et al., 1971
Rat/Wistar	Aroclor 1254	0, 6.25, 12.5, 25.0, 50, 100 mg/kg/day on days 6-15 of gestation	None reported	None reported	
Rat/Sherman	Aroclor 1254	0, 1, 5, 20, 100, 500 mg/kg diet from 3-4 weeks of age to termination of 1- or 2-generation study	None reported	F_{1a} 500 mg/kg: reduced litter size; 100 mg/kg: reduced litter size; F_{1b} 100 mg/kg: reduced survival at weaning; 20, 100 mg/kg: reduced litter size; F_{2a} 100 mg/kg: reduced litter size and reduced survival at weaning; 20 mg/kg: reduced litter size; F_{2b} 20, 100 mg/kg: reduced litter size.	Linder et al., 1974
	Aroclor 1260	0, 5, 20, 100, 500 mg/kg diet from 3-4 weeks of age to termination of 1- or 2-generation study	None reported	F_{1a} 500 mg/kg: reduced litter size and reduced survival at weaning. F_{1b} 500 mg/kg: reduced litter size.	
	Aroclor 1254	0, 10, 50, 100 mg/kg/day dosed on days 7-15 of gestation	None reported	F_1 100 mg/kg/day: reduced litter size.	
Rat/Sprague-Dawley	Aroclor 1254	0, 25, 50, 100, 200, 300, 600, 900 mg/kg diet/day on days 6-15 of pregnancy	≥600 mg/kg: partial anorexia and weight loss.	≥300 mg/kg: fetal death at delivery. ≥100 mg/kg: reduced litter weight.	Spencer, 1982
Mice/CD_1	3,3',4,4',5,5'-HCB	0, 0.1, 1.0, 2.0, 4.0, 8, 12 mg/kg/day, on days 6-15 of gestation	≥8 mg/kg/day: reduced rate of gain, lethargy, vaginal bleeding.	≥4 mg/kg/day: fetal mortality, resorptions; ≥2 mg/kg/day: increased incidence of cleft palate; ≥4 mg/kg/day: increased incidence of hydronephrosis; ≥1 mg/kg/day: increased incidence of cream colored liver; ≥1 mg/kg/day: increased incidence of undersized renal papillae.	Marks et al., 1981

TABLE V-21 (cont.)

Species/ Strain	Source of PCBs	Dosage Level and Duration of Treatment	Maternal Response	Progeny Response	Reference
Mice/ddY	Kanechlor 500	0, 0, 10, 20, 30, 40, 50 mg/mouse days 6-16 of gestation	Mortality	≥10 mg/mouse: cleft palate.	Watanabe and Sugahara, 1981
Rat/Sprague-Dawley	Aroclor 1221 1242 or 1260	0 or 30 mg/kg bw, days 14-20 of pregnancy	None	None	Gellert and Wilson, 1979
Rat/Wistar	Aroclor 1254	0 or 70 mg/ℓ drinking water (6.4 mg/kg bw); 9 weeks	Mortality at 7 weeks	Fetal resorption	Baker et al., 19
Mice/NMRI	Clophen A60	0 or 0.025 mg/day; 72-76 days	Lengthened estrus cycle	Fetal resorption	Orberg and Kihlstrom, 1973
Mice	2,2',4,4',5,5'-hexachlorobiphenal	0 or 0.5 mg/day; 7 or 13 days of pregnancy	Hepatomegaly	None	Mattson et al., 1981
Rabbit	Aroclor 1221, Aroclor 1254	0, 1.0 or 10 mg/kg bw; first 28 days of pregnancy	None	None	Villeneuve et al 1971
Mink	Aroclor 1242	0-40 mg/kg diet; 8 months	≥10 mg/kg: significantly increasing mortality; ≥5 mg/kg: complete reproductive failure;	NA NA	Bleavins et al., 1980
	Aroclor 1016	20 mg/kg diet	25% mortality, reduced fertility.	Increased mortality, decreased body weight by 4 weeks.	
Monkey/ cynomolgus	Aroclor 1254	0, 100 or 400 mg/kg bw/day; continuous starting at 60 days of pregnancy	Fingernail loss; immunologic incompetence.	Fetal mortality; immunologic incompetence.	Truelove et al., 1982
Monkey/ Rhesus	Aroclor 1248	0, 2.5 or 5.0 mg/kg diet; 18 months starting 6 months pregestational	Facial edema, cachexia, hair loss, hyperpigmentation	Fetal death, infant facial and eyelid edema, loss of facial hair, facial hyperpigmentation, gastric hyperplasia and vomiting, lumphoid degeneration, hypocellular bone marrow, fatty liver.	Allen and Barsotti, 1976; Allen et al., 1979b, 1980
	Aroclor 1248	0, 2.5 or 5.0 mg/kg diet; off treatment for 1 year	Persistent hyperpigmentation	Fetal death; reduced neonatal weight hyperpigmentation.	

NA - Not applicable

These workers found rabbits considerably more sensitive to Aroclor 1254 (Villeneuve et al., 1971). In three separate experiments, pregnant rabbits (breed or strain not given) were treated with Aroclor 1254 dissolved in corn oil at levels of 1, 10, 12.5, 25.0 or 50.0 mg/kg bw/day by gavage for the first 28 days of pregnancy. Combined numbers of rabbits/dosage level, respectively, from the three experiments were as follows: 16, 3, 4, 2, 8 and 4. An additional group of four rabbits was treated with 25 mg/kg Aroclor 1254 on days 7-28 of pregnancy. Rabbits were killed on the 29th day and uteri were examined. Examination of the uteri of 16 control animals revealed one dead fetus, 0 resorptions and 0 fetuses aborted. Dead fetuses, resorptions and abortions, respectively, for groups treated for the first 28 days of pregnancy were as follows, 1.0 mg/kg/day, 0, 1, 0; 10 mg/kg/day, 0, 0, 0; 12.5 mg/kg/day, 5, 6, 2; 25.0 mg/kg/day, 18, 9, 10; and 50.0 mg/kg, 12, 6, 3. Rabbits treated on days 7-28 of pregnancy with 25.0 mg/kg experienced 6, 0 and 6 dead fetuses, resorptions and abortions, respectively. Two fetuses from dams in the 12.5 mg/kg/day group had subcutaneous cephalic hemorrhages and cranial asymmetry. Maternal toxicity was manifested as death of two dams in the 50.0 mg/kg group and one dam in the 25.0 mg/kg group. Other evidence of maternal toxicity consisted of weight loss in dams receiving 25 or 50 mg/kg and hepatomegaly in dams receiving 10, 25 or 50 mg/kg. Dams treated with 25 mg/kg on days 7-28 of pregnancy exhibited decreased weight gain and hepatomegaly.

Another study in the same laboratory (Villeneuve and Grant, 1971) determined no evidence of fetal toxicity to orally administered Aroclor 1221 or Aroclor 1254 at levels of 0, 1.0 or 10 mg/kg bw in rabbits. Groups of four mature does of unspecified strain were exposed for the first 28 days of

Although no deaths occurred in exposed dams, lethargy and vaginal bleeding were observed in dams exposed to $\geq$8 mg/kg/day. Dams in the 8 and 16 mg/kg/day groups suffered significant ($p<0.05$) reduced weight gain compared with controls. Average numbers of live fetuses/dam in the low- to high-dose groups were 12.4, 12.4, 12.2, 12.0, 11.0, 9.9 and 9.8, respectively. These data were significantly ($p<0.01$) dose-related at or above the 4 mg/kg/day level when subjected to Jonckheere's test. Average number of implants was reduced ($p<0.05$) in the 8 mg/kg/day group and fetal resorptions were increased in the 8 and 16 mg/kg/day group. Possibly because of the greater care used in examining fetuses, terata were found in this study. Significant ($p<0.001$) incidences of cleft palate occurred in groups dosed at $\geq$2 mg/kg/day, apparently related to dosage level. Occurrence of cleft palate/numbers of fetuses examined were 2/247, 4/83, 5/63, 20/117, 69/154, 118/176 and 82/108 for the groups as described above. Hydronephrosis also followed a significant ($p<0.001$) dose-related trend with incidences of 2/247, 0/83, 1/63, 6/117, 25/154, 83/176 and 30/108, respectively. Significant variations in viscera were also noted. Cream-colored liver nodes were noted in 1, 0, 7, 21, 38, 46 and 24 fetuses of those examined in each group, respectively. These data were significant ($p<0.001$) at $\geq$1 mg/kg/day treatment level. Likewise, undersized renal papillae were observed in 0, 0, 6, 10, 27, 9 and 0 fetuses of those examined in each group, respectively. These data were significant ($p<0.001$) at the 2-8 mg/kg/day treatment levels. The authors mentioned briefly that they also found 3,3'4,4'tetrachlorobiphenyl to be teratogenic, but that HCB was the more potent PCB. Stating that they had tested several other isomers including all other possible isomers of hexachlorobiphenyl (unpublished data), the authors concluded that chlorine at the 3,3',4 and 4' positions are essential for teratogenicity.

pregnancy; then they were killed and the gravid uteri were examined. Nidation, fetal growth and development, litter size and placentation were all similar to control rabbits. The only evidence of maternal toxicity reported was an increase in dam liver weights in the 10 mg/kg Aroclor 1254 treatment group, but no actual data were given. The authors concluded that at these dosage levels (lower than those used in the previously reported study) there was no evidence of fetal toxicity caused by Aroclor 1221 or Aroclor 1254 in the rabbit.

In a more recent study, Marks et al. (1981) clearly demonstrated teratogenicity in CD-1 mice resulting from treatment with 3,3',4,4'5,5'hexachlorobiphenyl. This isomer was chosen for these studies because it was found to be more toxic and more strongly bioaccumulated, and to have a more pronounced effect on liver, thymus and spleen than other isomers tested (Broca et al., 1981). Pregnant mice were divided into seven groups and administered HCB in cottonseed oil at levels of 0.1, 1.0, 2.0, 4.0, 8 or 16 mg/kg/day by gavage. The control groups consisted of 63 mice; numbers in HCB groups ranged from 13-24. Treatment started on day 6 of gestation (vaginal plug day = day 1) and continued through day 15. On day 18 of gestation mice were killed and examined. Implantations were counted, the general condition of each conceptus was recorded and live fetuses were weighed, sexed internally and examined for external malformations. At least 33% of the fetuses from each litter and all "stunted" fetuses (weighing <0.5 g or <2/3 of mean weight of larger litter mates) were examined for visceral malformations. The heads of these fetuses were examined by free hand sectioning. All fetuses were subjected to skeletal examinations.

A commercial mixture of PCBs, Kanechlor 500, also induced formation of cleft palate in mice (Watanabe and Sugahara, 1981). Groups containing 24-119 pregnant ddY strain mice were injected subcutaneously with 95% ethanol containing 20-100 mg PCB/m. Amounts of PCB injected daily were 1.0, 2.0, 3.0, 4.0 or 5.0 mg/mouse/day. Apparently a vehicle control group was used as well as an untreated control group. Treatment began on day 6 and terminated on day 16 of gestation. Mice were killed on day 18 and examined. Maternal mortality was 0/119, 1/70, 0/65, 0/50, 1/45, 3/52 and 11/89 in the untreated control, vehicle control, 10, 20, 30, 40 and 50 mg/mouse (total doses) PCB treated groups, respectively. In these same groups, the incidence and percent of dead and resorbed fetuses was 63/1197 (5.3%), 35/666 (5.3%), 37/551 (6.7%), 28/446 (6.3%), 27/280 (9.6%), 34/341 (10.0%) and 24/249 (9.6%), respectively. These data were not subjected to statistical analysis. The incidence of cleft palate in live fetuses in the respective groups, 0/1134 (0.0%), 0/631 (0.0%), 1/514 (0.2%), 4/418 (1.0%), 9/253 (3.6%), 12/307 (3.9%) and 13/225 (5.8%) was apparently dose-related and significant ($p<0.05$). Additional experiments were performed on two other groups of mice. One group received 5/mg/day Kanechlor 500 on days 6-10 of gestation (total 25 mg/mouse), and one group received 10 daily doses of 5 mg Kaneclor 500/day before mating (50 mg total) and an additional 10 daily doses during days 6-16 of gestation (100 mg total/mouse). There was no mention of the use of controls in these trials. Severe mortality (6/22) was observed in the 100 mg total group. Resorbed or dead fetuses and cleft palate in live fetuses occurred in the 25 total (7/93 and 1/86, respectively, and 100 mg total (6/34 and 5/28, respectively) groups. These workers also reported five instances of brachydactyly, four of syndactyly and one of cleft lip in PCB-treated fetuses.

An extensive study on reproductive effects of PCBs in rats was performed by Linder et al. (1974). A preliminary 1-generation study was followed by a 2-generation study where Aroclor 1254 or Aroclor 1260 was fed in the diet continuously starting at 3-4 weeks of age. In the 1-generation study, groups of 10 specific pathogen-free female Sherman rats were fed diets containing either Aroclor 1254 or Aroclor 1260 at levels of 0, 100 or 500 mg/kg/day starting at 3-4 weeks of age and terminating after the weaning of the F_{1b} litter. In the 2-generation study, groups of 10 specific pathogen-free Sherman rats of each sex were fed diets levels of 0, 1, 5, 20 or 100 mg/kg Aroclor 1254 or 0, 5, 20 or 100 mg/kg Aroclor 1260. All F_1 offspring from these groups were started on their respective PCB-containing diet at weaning, ~3 weeks of age. In a post-implantation exposure study, 100-day-old stock rats were treated with Aroclor 1254 in peanut oil by gavage at levels of 0, 10, 50 or 100 mg/kg bw/day or Aroclor 1260 by gavage at levels of 0 or 100 mg/kg bw/day on days 7-15 of pregnancy. The Aroclor 1254 study groups consisted of 9 or 10 rats; the Aroclor 1260 study groups consisted of 12 rats each.

Aroclor 1254 at the 500 mg/kg level resulted in reduction of number of litters born (4/10) compared with controls (8/10) (Linder et al., 1974) Litter size (4.0) was reduced ($p<0.05$) compared with controls (11.1) at birth. Of these litters, 2/4 contained no live offspring; all offspring from the remaining two litters died by postpartum day 3. In the 2-generation study, F_{1a} litters were reduced in size (10.7) compared with controls (12.4) at the 100 mg/kg level of Aroclor 1254 ($p<0.05$). F_{1b} litters were reduced in size (10.4, $P<0.05$; 9.5, $p<0.005$) compared with controls (12.3) at the 20 and 100 mg/kg level, respectively. Size of F_{2a} and F_{2b}

litters for Aroclor 1254 levels of 0, 20 and 100 mg/kg were 12.5, 10.6, 7.2 and 12.7, 9.6, 3.5, respectively. These reductions in litter size were significant at the 0.5% confidence level with the exception of the F_{2a} 20 mg/kg litter, which was significant at the 5.0% confidence level. Percent weaned was significantly ($p<0.005$) reduced in F_{1b} litters (68.1%) compared with F_{1b} controls (100%) at the 100 mg/kg treatment level in the 1-generation study. F_{2a} litters experienced a significant ($p<0.005$) decrease in percent weaned at the 100 mg/kg treatment level (77.8%) compared with controls (97.8%).

Aroclor 1260 appeared to cause a significant ($p<0.001$) decrease in litter size in F_{1a} rats (8.5) compared with controls (12.3) and F_{1b} rats (6.7) compared with controls (11.0) at the 500 mg/kg level in the 1-generation study (Linder et al., 1974). F_{1a} survival (38.2%) was also significantly reduced at the 500 mg/kg level compared with controls (99.1%). Aroclor 1260 failed to produce evidence of reproductive toxicity at levels <100 mg/kg.

Significant ($p<0.001$) effects on reproduction in the post-nidation study were found only at the highest (100 mg/kg/day) treatment level of Aroclor 1254 (Linder et al., 1974). Survival at weaning (30.1%) was reduced compared with controls (98.2%). No effects from Aroclor 1260 were noted.

In none of these studies was any evidence of maternal toxicity mentioned (Linder et al., 1974). Upon necropsy, F_0 males and females of many Aroclor-treated groups exhibited hepatomegaly and reduced PCV and hemoglobin. These and other effects have been discussed in the Chronic and Subchronic Toxicity Section.

Recently, Spencer (1982) investigated the reproductive toxicity of Aroclor 1254 in female Sprague-Dawley rats. Groups of seven rats were fed diets containing Aroclor 1254 at levels of 25, 50, 100, 200, 300, 600 or 900 mg/kg diet from days 6-15 of pregnancy. Implantations were counted at days 6 and 12 of pregnancy. Dietary levels of 300, 600 or 900 mg/kg led to daily feed intakes of 15.8, 9.6 or 7.0 g/rat/day, significantly ($p<0.05$) less than controls (20.8 g/rat/day). No effect on the number of implantations observed on days 6 or 12 of pregnancy was noted. Average fetal weight/litter at birth was significantly ($p<0.05$) reduced at the 100 (6.19 g), 200 (5.61 g) and 300 (5.58 g) mg/kg level compared with controls (7.02 g). Fetal toxicity at birth in the higher dosage groups precluded these measurements. Percentage live births/litter was significantly ($p<0.05$) depressed at the 300 (28.03%) and 600 (17.14%) mg/kg level compared with controls (79.54%). There were no live deliveries at the 900 mg/kg level. There was no mention of maternal toxicity other than the severely depressed food intake previously discussed and the resultant weight loss during treatment of the 600 and 900 mg/kg groups. The author suggested that poor nutrition resulting from depressed food intake may have exacerbated the toxic effects of Aroclor 1254.

Using a low level (30 mg/kg bw) of Aroclors 1221, 1242 and 1260 and various chlorinated pesticides, Gellert and Wilson (1979) investigated the effects of prenatal exposure on future reproductive functions of F_1 males and females. Speculating that PCBs may have estrogenic qualitites that could be expected to interfere with subsequent reproduction, groups of 25-37 Sprague-Dawley rats were treated by gavage on days 14-20 of pregnancy.

There was no mention of maternal or fetal toxicity. At 6 months of age, F_1 females were examined for persistent vaginal estrus and anovulation, indicators of prenatal exposure to estrogenic substances. Although no significant increase in these parameters was found, the incidence seemed to be inversely related to the degree of chlorine saturation of the Aroclor tested. Male F_1 rats at 6 months of age were mated with known stock breeder females and then necropsied. The offspring were normal, and examination of adrenals, testes, epididymus and ventral prostates of the F_1 males revealed no lesions.

An earlier study (Dikshith et al., 1975) also examined the effects of Aroclor 1254 on rat testis. Groups of 18 mature male Sprague-Dawley rats were treated by gavage for 7 consecutive days with corn oil (vehicle control) or 50 mg/kg bw Aroclor 1254. At the end of the 7-day treatment period, the rats were allowed a period of recovery. No signs of morbidity were observed. Necropsy examinations were performed on three rats from each group on days 1, 7, 15 and 30 days of recovery. Body weight of rats examined by necropsy at 30 days (374.5 g) was significantly ($p<0.05$) reduced compared with controls (474.0 g). Testicular and epididymal size and histological features were normal in treated rats with the exception of a slight increase in testicular interstitial tissue exhibiting increased acid phosphatase activity. The authors suggested that this may indicate enhanced secretion of androgens.

One recent study on early postnatally-administered PCBs to male rats was undertaken to determine effects on their subsequent reproductive performance (Sager, 1981). At 1 day of age, litters of Holtzman rats were reconstituted to contain 10 males and treatment was begun. A group of 6 dams and litters were given peanut oil and served as controls. Treatment groups consisted of 6 dams receiving 8 mg/kg bw, 6 dams receiving 32 mg/kg bw and 7 dams receiving 64 mg/kg bw Aroclor 1254 dissolved in peanut oil. Administration was by gavage on days 1-3, 5 and 7 of lactation. At 130 days of age, 3 or 4 males from each group were randomly selected for observation on mating behavior and fertility. Males were subsequently killed and examined for organ weights: liver, ventral prostate, seminal vesicles and testis. Interference with mating behavior and fertility was evidenced by a reduction in numbers of females impregnated [10/10, 8/9, 7/10 ($p<0.05$), and 6/8 ($p<0.05$) in the 0, 8, 32 and 64 mg/kgtreated groups, respectively]. Testicular weight (g)/percent body weight for the 0, 8, 32 and 64 mg/kg-treated groups were 3.87/0.831, 4.09/0.891, 4.25/1.008 and 4.36/1.010, respectively. These data were significant ($p<0.05$) in the 32 and 64 mg/kg-treated groups. The author reported a decrease in ventral prostate and seminal vesicle weight directly related to dosage level. The author reasoned that accessory sex gland development requires adequate androgenic stimulation and that testiculomegaly reflects excessive gonadotropic stimulation because of reduced androgenic feedback by the hypothalamicpituitary axis. Testiculomegaly and subnormal development of accessory sex glands was taken as evidence that PCBs administered in the early postnatal period interfered with circulating levels of androgens. Under the conditions of this study, Aroclor 1254 exposure during the early postnatal period interfered with subsequent male fertility.

In an effort to demonstrate that PCBs may induce microsomal hydroxylating enzymes which catalyze the breakdown of steroid hormones, Orberg and Kihlstrom (1973) investigated the ability of Clophen A60 to affect reproduction in mice. NMRI-strain sexually mature female mice were fed 0.025 mg/day Clophen A60 in peanut oil (23 mice) or peanut oil alone (22 control mice) for 72-76 days. Estrus cycle length was determined by examination of vaginal smears of 11 control and 14 treatment mice for a 62-day period. All 45 mice were mated after 62 days of feeding and were examined for number of corpora lutea and implantations on days 8-10 of pregnancy. Clophen A60 was found to lengthen the estrus cycle ($p<0.0005$) from 6.6-8.7 days and to reduce the rate of nidation ($p<0.025$) from 87.0-79.5%.

In a subsequent study, Orberg (1977) failed to demonstrate any effect of pre- and postnatally-administered PCBs on the reproductive performance of mice. TCB or HCB dissolved in peanut oil was fed at levels of 0.05 mg/day from day 5 of pregnancy to weaning at postpartum day 22 to NMRI-strain mice. A third group consisted of peanut oil-fed controls; 35 pregnant mice were used in this study. On the day of birth, litters were reconstituted to contain 10 young/litter. Since this study investigated the effect of PCBs on reproductive performance of the progeny of treated dams, no data on the parameters of reproductive performance of treated dams were given. At weaning, male and female F_1 progeny were separated and exposure to PCBs was terminated. When sexually mature, one male was caged with one female until the presence of a vaginal plug indicated pregnancy. The males and females thus mated had each been exposed to the same PCB through their dams. Pregnant females from these matings were killed and fetuses were counted. The numbers of F_1 mice found pregnant from dams in the control, TCB- and HCB-exposed groups were 43/51 (84.3%), 29/43 (67.4%) and 36/48 (75.0%), respectively. Numbers of fetuses/pregnant female were 10.8, 11.6 and 11.1, respectively. These differences were not significant. In companion studies (Chapter III), the author determined that both maternal tissues and milk and fetal tissues bioaccumulated higher levels of HCB than PCB when dams were exposed to 0.05 mg/day of either PCB during days 5-19 of pregnancy. Orberg (1978) further investigated the effects of TCB and HCB fed at two different dosage levels on reproduction in mice. About 230 NMRI mice were divided into five groups on the day in which a vaginal plug was first found. These groups were fed peanut oil (controls) or peanut oil containing 0.05 or 0.5 mg TCB/day, or 0.05 or 0.50 mg HCB/day beginning on the first day of vaginal plug for 6 consecutive days. On the fourth day of exposure, 11-12 mice from each group were killed for liver enzyme studies. The remaining females were killed on the day after last exposure and were examined for numbers of corpora lutea and placentae. Numbers of pregnancies/group and average numbers of corpora lutea/mouse for the control, low- and high-dose TCB and low- and high-dose HCB groups were 40/56 (71.4%), 11.33; 15/20 (75.0%), 10.93; 28/40 (70.0%), 12.04; 14/20 (70.0%), 11.64; and 24/32 (75.0%), 11.83, respectively. These differences were not significant. The average numbers of placentae, interpreted as implantations, in the five groups were 10.45 (92.3%), 10.13 (92.7%), 10.57 (87.8%), 11.00 (94.5%), and 9.92 (83.8%), respectively. Significant ($p<0.001$) differences from control data were found in the TCB 0.50 mg/day and HCB 0.50 mg/day exposed mice. The author concluded that under the conditions of this study no differences in fetotoxicity were detected between these two PCBs, which differ considerably in chlorine content.

Baker et al. (1977) administered Aroclor 1254 to rats through drinking water to study the toxicokinetics of this commercial mixture of PCBs (Chapter III). Because the effects of Aroclor 1254 on reproductive function were reported, this study is reviewed here. Drinking water containing 70 ppm Aroclor 1254 emulsified with 0.15% Tween 80 or drinking water containing 0.15% Tween 80 was given to 250 mature (344±40 g) male and 250 mature (220±34 g) female Wistar rats randomly separated into matched experimental and control groups. The authors stated that Aroclor 1254 consumption was 6.4 mg/kg bw but did not report how this figure was obtained. Treatment was continued for 9 weeks, at which time it became evident that fetal resorption was occurring and all groups were returned to plain tap water. Each matched group of 125 rats was further subdivided into 25 subgroups of five rats each. Each week, two males from one subgroup were placed in the cage with a subgroup of five females. After 2 weeks, these males were removed and placed with stock female rats to determine male fertility and reproductive success. This procedure was done for each of the 25 subgroups of control and treatment male and female rats. No adverse effects on male fertility were reported; litter size of PCB-exposed males (8.2±3.1) was similar to control males (8.3±3.4) when mated with control females. Although body weight remained similar among control and treatment groups, "several" females exposed to PCBs died during the 7th week. Necropsy revealed no gross lesions. Although the authors stated that fetal resorption was obvious during the 8th and 9th week of exposure, data were not reported to support this assertion.

In a study designed to evaluate the effects of the PCB 2,2'4,4'5,5'hexachlorobiphenyl, Mattson et al. (1981) were unable to increase fetal resorption in CBA mice mated syngeneically to CBA or allogeneically to NMRI male mice. Groups of 10 female mice were fed 0.1 mℓ peanut oil containing 0 or 0.5 mg PCB once daily from day 5-11 of gestation. In a second trial, groups of 10 females were treated with peanut oil containing 0 or 0.5 mg PCB from day 5-18 of pregnancy. All mice were killed the day following last treatment except for five control and five treated mice from the second trial, which were allowed to give birth to permit examination of offspring. Treatment with PCB at this level did not result in significant increase in fetal resorption. Interestingly, control mice allowed to deliver either resorbed or killed their litters, while PCB-treated mice delivered "normal" litters, precluding further studies on reproduction. The authors speculated that PCB-treated mice tolerated a move to a different animal room with noisy fans better than control mice. They felt that PCB-treated mice seemed to be less sensitive to any kind of disturbance than control mice.

Using a low chlorinated PCB, 2,2'-dichlorobiphenyl, Torok (1978) was able to increase fetal absorption and retard fetal development. Pregnant NMRI mice were orally dosed with 375, 500 or 750 mg/kg of this PCB daily for 3 days following appearance of the vaginal plug. Another group was orally dosed with 250 mg PCB/kg/day for 6 days following appearance of the vaginal plug. Further details of experimental protocol were lacking in the abstract of this study. The author reported that the 375 mg/kg dose increased fetal resorptions and that doses ≥500 mg/kg caused delayed implantation.

Effects in Other Species -- It has been discovered that mink are especially sensitive to the toxic effects of PCBs. Lake Michigan coho salmon and other fish may bioaccumulate levels of PCBs that, when fed to mink, are sufficient to precipitate acute and chronic toxicity. One laboratory (Ringer et al., 1972; Aulerich et al., 1973; Aulerich and Ringer, 1977; Bleavins et al., 1980) has investigated this phenomenon. By feeding diets containing Lake Michigan fish canning wastes or various Aroclor products, they were able to demonstrate comparable tissue levels of PCBs (Aulerich and Ringer, 1977; Aulerich et al., 1973). They concluded that PCBs were the toxic agents and undertook an 8-month feeding trial with Aroclors 1016 and 1242 to determine reproductive toxicity (Bleavins et al., 1980). Levels of Aroclor 1242 as low as 10 ppm diet resulted in heavy mortality and 5 ppm resulted in complete reproductive failure. Aroclor 1016 at 20 ppm caused 25% mortality and reduced fertility of females. By 4 weeks, litters experienced increased mortality (not statistically significant) and decreased litter biomass ($p<0.05$). No terata were observed. These workers concluded that mink are extremely sensitive to PCBs and that the more heavily chlorine substituted isomers are more toxic. Furthermore, reproductive toxicity was related to fetotoxicity rather than interference with ovulation or nidation because fetal resorptions were found at all stages of gestation.

A comparison of the fetotoxic effects of Clophen A50 and 2,2'4,4'5,5'-hexachlorobiphenyl (HCB) in guinea pigs was performed by Brunstroem et al. (1982) in three separate trials. In the first trial, groups of 19 or 18 pregnant Dunkin Harley guinea pigs were exposed to 2.2 mg/day Clophen A50 in peanut oil by oral administration for days 16-60 of gestation. Exposed animals received a total dose of 100 mg. In the second trial, groups of 15 or 16 guinea pigs were exposed to a total dose of 0 or 25 mg HCB distributed over days 16-60 of gestation. The third trial involved exposing 5 or 19 guinea pigs to a total of 0 or 100 mg HCB over days 22-60 of gestation. On day 61 of gestation, laparotomy was performed and uteri were examined for resorptions and live and dead fetuses. All dams seemed unaffected by treatment. Macerated fetuses were aborted between day 50 and 55 by four dams in the Clophen A50 group. Upon laparotomy, the remaining 14 dams were found to contain 14 live and 31 dead fetuses, significantly ($p<0.0005$) different from the 19 control dams that contained 65 live fetuses and 1 dead fetus. No abortions occurred in the other treatment or control groups. Numbers of live and dead fetuses in the control and 25 mg HCB and control and 100 mg HCB-treated groups were 47, 1; 47, 4 and 19, 0; 69, 4, respectively. These data were not significant. According to the authors, the HCB used in these trials contained <0.5 mg/kg PCDFs. They suggested that PCBs other than HCB or possibly PCDFs or other contaminants in Clophen A50 accounted for the fetotoxicity to guinea pigs observed in these studies.

Hansen et al. (1975) investigated the effects of PCBs on swine reproduction. Groups of 5 third-litter sows of mixed breeding were fed rations containing 0 or 20 ppm Arolcor 1242 throughout gestation and nursing. Sows were allowed to complete at least one estrus cycle on their respective rations before being mated. All five control sows conceived and farrowed 11.4 ± 1.14 live pigs/litter; only four PCB-exposed sows conceived and their litters averaged 6.4 ± 4.39 live pigs ($p<0.05$). It appeared that PCB-exposed sows delivered more mummified fetuses, but these results were not significant. Satisfactory control of experimental variables in protocols using farm animals is extremely difficult. All that can be concluded from this study is that exposure to Aroclor 1242 seemed to reduce the number of live pigs delivered/litter.

Two laboratories reported studies on PCB toxicity in subhuman primates. Truelove et al. (1982) dosed 4, 60-day pregnant cynomolgus monkeys with 100, 100 or 400 μg/day Aroclor 1254 continuously until infants were stillborn or died. Both monkeys dosed at the 100 μg/day level delivered term, stillborn infants. The monkey dosed at the 400 μg/day level delivered a live infant that subsequently succumbed at 139 days of age to pneumonia, which these workers judged to result from impaired immunocompetency that might be a toxic manifestation of PCBs. Maternal toxicity was evidenced as fingernail loss and impaired immunocompetence as evaluated by titer developed to tetanus toxoid and sheep erythrocytes.

A longer-term and more intensive study examined the effects of Aroclor 1248 on reproduction in rhesus monkeys (Allen and Barsotti, 1976; Allen et al., 1979b; Allen et al., 1980). Aroclor 1248 was fed to groups of eight mature female monkeys weighing ~5.6 kg at levels of 2.5 or 5.0 ppm diet for ~18 months starting 6 months before breeding to untreated males. On the basis of food intake, the authors calculated total Aroclor 1248 intake to be 270±25 and 498±50 mg for the 2.5 and 5.0 mg/kg diet groups, respectively. Analysis of this batch of Aroclor 1248 revealed ~1.7 mg/kg PCDFs. This would have provided 4.4 and 8.7 μg/kg PCDFs in the 2.5 and 5.0 mg/kg Aroclor 1248 diets, respectively. Treated females experienced lengthened menstrual cycles accompanied by prolonged and lower levels of serum progesterone. Conceptions were 8/8, 8/8 and 6/8 for 2.5 and 5.0 mg/kg, respectively. All control animals delivered full-term live infants, monkeys receiving 2.5 mg/kg delivered five live infants and experienced three early abortions or fetal resorptions, and only one monkey receiving 5.0 mg/kg delivered a live infant. Treated dams exhibited cachexia, facial edema, hyperpigmentation and hair loss. Live infants had reduced birth weights and, after 2 months of nursing, exhibited facial edema, hyperpigmentation and hair loss and palpebral edema and acneform lesions more severely than their dams. Within the first year, 3 of these 6 infants had died. These workers presumed that PCB transmission through lactation, causing infant tissue levels of PCBs to exceed maternal levels, caused this early infant loss. Removing treated monkeys from treatment for 1 year allowed them to recoup body weight and permitted facial lesions to heal. Hyperpigmentation, however, persisted. These monkeys were then rebred to control males. Menstrual cycles had returned to normal. Pregnancies resulting were 8/8 for monkeys that had been fed both 2.5 and 5.0 mg/kg. Full-term live infants resulting were 6/8 and 7/8, respectively, and they weighed less than infants born to control animals. Reproductive statistics on control animals were not given. After nursing for 4 months, infants from previously exposed dams evidenced focal areas of hyperpigmentation.

In a brief abstract of this work in progress, Barsotti and Allen (1975) reported that by 52 weeks, females on the 2.5 and 5.0 mg/kg diet had received 182 and 364 mg Aroclor 1248, respectively. Males were fed the higher level only and had received 383 mg Aroclor 1248. After 6 months, the conception rate of control females bred to PCB-treated males was similar to the conception rate of control females bred to control males. These workers concluded that under the conditions of this study, exposure to PCBs for 6 months at the levels given does not interfere with male fertility.

subsequent report from this laboratory (Allen et al., 1980) indicated
the estrus cycle returned to normal and that fertility improved in the
ed females 1 year after being returned to control (recovery) diet.
1-5 breedings, 8/8 females from the 2.5 mg/kg diet and 7/7 females
the 5.0 mg/kg diet conceived. These pregnancies resulted in 7 and 4
l births in the 2.5 and 5.0 mg/kg diet groups, respectively. These
s, on the basis of recorded feed intake, were credited with having
ved a total of 243-309 mg and 460-614 mg of Aroclor 1248, respec-
y. All eight control animals conceived and each delivered a live
t. Birth weights were 516±65, 480±83 and 440±55 g ($p<0.05$) for con-
2.5 and 5.0 mg/kg diet groups, respectively. Signs of toxicity
ing anorexia, weight loss, swollen eyelids, alopecia, acne and devel-
t of scaly skin in nursing infants. Death of two infants in each
ent group occurred before weaning.

a brief abstract (Barsotti, 1981) of the continuation of these
s, it was stated that two other breeding trials during the recovery
resulted in reduced infant production. No information was available
ning the length of recovery period, number of females mated from each
or number of matings/female allowed.

is same abstract (Barsotti, 1981) summarized a study of feeding diets
ning 0, 0.025, 0.25 or 1.0 mg/kg Aroclor 1016 to 24 adult female
monkeys. The length of exposure time was not given. No abnormali-
f clinical or reproductive parameters were noted. Birth weights of
born to the 1.0 mg/kg diet group were significantly ($p<0.01$) less
at of control animals, but actual data were not given.

Reproduced from
best available copy.

the
CBs,
ina-
-ICR-
the
the
PCBs
post-
ovide
e six
PCB-
ntain
spring
urred;
end of
eighed
sed to
ing at
group
PCB-0.4
mercury
ight of
Subse-
eeks of
-exposed
or male

fertility. These matings included 9-23 females from each group. Reproductive studies on 50% of these females continued for up to 48 weeks. No significant differences in reproductive efficiency (live births/female/year) or litter size were noted. Significant levels of terata were not found in any offspring examined.

Summary

Single oral LD_{50}s reported in rats ranged from 1.01 g/kg bw (Aroclor 1254) (Garthoff et al., 1981) to 19.2 g/kg bw (Aroclor 5460) (Fishbein, 1974). Little variation in toxicity occurred by dividing the dose into five multiple doses (Garthoff et al., 1981). A single i.v. LD_{50} in rats of 358 mg/kg bw of Aroclor 1254 was reported (Linder et al., 1974). Death occurred in 5-110 minutes. Rats exhibited dyspnea, depression, salivation and diarrhea. Bleavins et al. (1980) reported an LD_{50} of 315-833 mg/kg bw (total dose) in minks maintained on diets containing Aroclor 1242 or 1016 for ~3 months.

Animals appeared to be slightly more sensitive to some pure isomers, with single-dose oral LD_{50}s in mice ranging from 19 mg of 3,4,5,3',4',5'-HCB/kg bw (Biocca et al., 1981) to 7.86 g of 2,4-DCB/kg bw (Kimbrough et al., 1978). Guinea pigs seemed most sensitive, with a reported LD_{50} of 0.5 mg/kg for 3,4,5-sym. HCB (McConnell and Kinney, 1978).

An early manifestation of acute toxicity in the rat was depressed feed and water intake and decreased body temperature (Komives, 1979; Komives and Alayoku, 1980). Increased liver weight after 7 days of exposure was a frequent finding in rats exposed to 12.5 mg Aroclor 1254/kg bw/day (Grant and fertility. These matings included 9-23 females from each group. Reproductive studies on 50% of these females continued for up to 48 weeks. No significant differences in reproductive efficiency (live births/female/year) or litter size were noted. Significant levels of terata were not found in any offspring examined.

Summary

Single oral LD_{50}s reported in rats ranged from 1.01 g/kg bw (Aroclor 1254) (Garthoff et al., 1981) to 19.2 g/kg bw (Aroclor 5460) (Fishbein, 1974). Little variation in toxicity occurred by dividing the dose into five multiple doses (Garthoff et al., 1981). A single i.v. LD_{50} in rats of 358 mg/kg bw of Aroclor 1254 was reported (Linder et al., 1974). Death occurred in 5-110 minutes. Rats exhibited dyspnea, depression, salivation and diarrhea. Bleavins et al. (1980) reported an LD_{50} of 315-833 mg/kg bw (total dose) in minks maintained on diets containing Aroclor 1242 or 1016 for ~3 months.

Animals appeared to be slightly more sensitive to some pure isomers, with single-dose oral LD_{50}s in mice ranging from 19 mg of 3,4,5,3',4',5'-HCB/kg bw (Biocca et al., 1981) to 7.86 g of 2,4-DCB/kg bw (Kimbrough et al., 1978). Guinea pigs seemed most sensitive, with a reported LD_{50} of 0.5 mg/kg for 3,4,5-sym. HCB (McConnell and Kinney, 1978).

An early manifestation of acute toxicity in the rat was depressed feed and water intake and decreased body temperature (Komives, 1979; Komines and Alayoku, 1980). Increased liver weight after 7 days of exposure was a frequent finding in rats exposed to 12.5 mg Aroclor 1254/kg bw/day (Grant and

Phillips, 1974). As little as 2 mg Clophen A-50/kg bw for 6 weeks resulted in increased liver size, congestion, fatty degeneration and focal necrosis. Apparently, considerable ultrastructural damage (intracellular vacuolization, fragmentation of rough endoplasmic reticulum and altered mitochondrial conformations) can result from one gavage treatment of 1.7 g 2,5,2',5'-TCB/kg bw without causing increased liver size (Lin et al., 1979). PCBs have been used widely to induce hepatic enzymes, often in studies with other chemicals. For that reason, larger doses are often given by injection or gavage to obtain a maximal response. Few studies identify threshold oral doses. Chu et al. (1977) and Garthoff et al. (1977) reported that 5-50 mg Aroclor 1254/kg diet for 2 weeks resulted in induction of many hepatic enzyme systems.

PCBs have been shown to cause splenic, thymic and lymph node regressions (Allen et al., 1975; Allen and Abrahamson, 1973; Parkinson et al., 1983), and considerable changes in the thyroid gland starting with enlargement and progressing to degeneration (Collins and Capen, 1980). The results of alterations in these organs have been pronounced immunosuppression (Allen et al., 1975) and reduction in serum thyroxin (Collins and Capen, 1980 a,b,c).

In general, hematologic values remained unchanged (Oishi et al., 1978) but blood and urine chemistries were affected by oral administration of PCBs. Blood levels of glucose were reduced and blood levels of urea nitrogen, cholesterol and protein were increased by Aroclor 1254 (Garthoff et al., 1977). Urinary levels of porphyrin and porphyrin precursors in the urine were noted following exposure to Clophen A-50.

Effects of PCBs on mice are similar to those in rats. Depressed food intake, lethargy and weight loss or reduced rate of weight gain were reported (Tanimura et al., 1980; Sanders et al., 1974; Marks et al., 1981). Damage to the liver in mice is similar to that observed in rats (Carter and Cameron, 1977; Orberg, 1978; Sanders et al., 1974; Loose et al., 1978a,b) and included enlargement, fatty degeneration and formation of microabscesses.

Mice have been used as a model to demonstrate the immunosuppressive effects of PCBs (Loose et al., 1978a,b). Reductions in circulating levels of antibodies to injected sheep erythrocytes or tetanus toxoid and reduced wheal formation to intradermal tuberculin have been reported. More importantly, increased mortality following PCB exposure was observed in mice injected with S. typhosa endotoxin or live malarial organisms, P. berghei (Loose et al., 1978a,b), or live S. typhimurium (Thomas and Hinsdill, 1978).

Manifestations of PCB toxicity in the rabbit resembled those in other species (Villeneuve et al., 1971) except that the rabbit may be more susceptible to kidney damage (Vos and Beems, 1971). Guinea pigs also exhibit many of the same signs of PCB toxicity as other species. Effects on the immune system of guinea pigs may be more profound than in other species (Vos and van Genderen, 1973).

Monkeys exhibit the same signs of PCB-induced toxicity as other species, with a few striking exceptions. Skin lesions such as edema of the face and eyelids are an early sign of toxicity induced by Aroclors (Allen et al., 1974a; Allen, 1975). These signs progressed to facial alopecia and development of comedones and eventually to a generalized edema and alopecia accompanied by pruritis. These authors also reported the development of gastritis, which progressed to a moderately invasive gastric hyperplasia. Accumulation of mucous in stomach glandular cells with consequent formation of large, mucus-containing cysts and glandular cell invasion of the muscularis was observed.

The signs of PCB chronic toxicity resemble closely the signs of acute toxicity. In rats, Morgan et al. (1981) reported alopecia, facial edema, exophthalmos, cyanosis and amber-colored urine after 72 weeks of oral exposure, and reduced body weight and stomach lesions such as intestinal metaphasia and adenocarcinoma after 104 weeks of exposure. Another laboratory (Allen et al., 1976; Allen and Abrahamson, 1979) reported normal hematologic parameters, hemoglobin, hematocrit and leukocyte count in rats after 52 weeks of exposure. Total serum lipids and cholesterol remained elevated throughout the study. Most dramatic alterations occurred in the liver. Hepatic RNA, protein and lipid content remained increased and DNA values were decreased throughout the study. Many microsomal enzymes were induced; glucose-6-phosphatase activity was inhibited. Jonsson et al. (1981) reported vascular engorgement in many organs of PCB-exposed rats. Ultrastructural examination of the liver revealed dramatic changes in hepatocellular cytoplasm, particularly in the endoplasmic reticulum.

Lesions in chronically-exposed mice closely resemble those in rats, but there seemed to be more mention of bile duct proliferations and oval cell formation (Ito et al., 1973). Amyloidosis, apparently inversely related to both the extent of chlorination and amount of material fed, was reported in this species (Ito et al., 1973). Other authors, however, made no mention of amyloidosis (Kimbrough and Linder, 1974). Nishizumi (1970) and Bell (1983) both reported eczematous lesions about the head and face.

Effects of chronic exposure of monkeys to PCBs also resemble those of acute exposure. Infants that had been exposed *in utero* and as nursing infants to Aroclor 1248 showed a progressive hyperactivity to 24 months of age and a retarded learning ability (Barsotti and Allen, 1975; Allen and Barsotti, 1976). At 44 months of age, the PCB-exposed infant monkeys exhibited hypoactivity (Bowman and Heironomus, 1981).

Chronic exposure to PCBs resulted in severe changes in the skin of monkeys. McNulty et al. (1980) reported a squamous metaplasia of sebaceous glands of the eyelids. Plugging the pores of these glands resulted in cystic degeneration of the meibomian glands. The nail beds had become hyperkeratotic, nails were elevated by accumulations of keratin, and many were shortened or lost. These authors also reported that exposed subjects had nearly complete regression of the thymus. The gastric mucosa was severely disrupted with loss of parietal and zymogenic cells and development of large mucus-containing cysts. In a second experiment, monkeys were exposed to 3,4,3',4'-TCB, 2,5,2',5'-TCB or Aroclor 1242. Those exposed to 3,4,3',4'-TCD exhibited the skin lesions described above, including squamous metaplasia of the meibomian glands, indicating that PCDFs may not be necessary to produce these lesions in monkeys.

Clearly, the liver is the primary target organ of PCB toxicity. Effects in rats (Allen and Abrahamson, 1979, Jonsson et al., 1981, Kimbrough et al., 1972), mice (Ito et al., 1973; Koller, 1977) and monkeys (Barsotti et al., 1976; Allen et al., 1979; Hori et al., 1982) are quite similar. The most detailed ultrastructural study was performed by Jonsson et al. (1981) in rats. These authors demonstrated that hepatotoxicity probably begins with venous engorgement, marked focal necrosis and attempts at regeneration. Hepatomegaly results from enlargement of individual hepatic cells, which is a consequence of lipid accumulation and greatly proliferated smooth endoplasmic reticulum. Proliferation of smooth endoplasmic reticulum is associated with induction of many hepatic microsomal enzyme systems (Bruckner et al., 1974a; Allen and Abrahamson, 1979). These factors probably contributed to the alterations reported in blood chemistries such as elevated serum cholesterol and lipids (Zinkl, 1977; Allen et al., 1976). Liver damage probably also resulted in elevated urinary porphyrin and prophyrin precursors (Bruckner et al., 1974b).

The immune system also seems to be a target of PCB toxicity. This phenomenon has been studied in most detail in mice. Loose et al. (1978a,b) found reduced levels of circulating antibodies in PCB-exposed mice compared with controls when immunized with sheep erythrocytes. They also reported greater mortality in PCB-exposed mice compared with controls challenged with _S_. _typhosa_ endotoxin or live _P_. _berghei_ organisms. Thomas and Hinsdill (1978) observed greater mortality in PCB-exposed mice than control mice when challenged with live _S_. _typhimurium_. Loose et al. (1978a,b) concluded that depressed immune function resulted before histologically discernible damage had occurred to thymus or spleen cells, and was therefore a sensitive indicator of PCB toxicity.

The thyroid gland seems to be another target of PCB toxicity. It has been studied most thoroughly in the rat by one laboratory (Collins et al., 1977; Collins and Capen, 1980a). These authors reported that rats exposed to PCBs experienced enlarged thyroids with reduced follicular size. Ultrastructural examinations revealed that follicular cells were hypertrophied and filled with colloid. These authors concluded that the intoxicated follicular cells were unable to clean the colloid and secrete thyroxin. Serum thyroxin levels thus dropped. In a sophisticated protocol using brown rats (known for their inability to conjugate, hence eliminate, thyroxin) these authors (Collins and Capen, 1980a) confirmed that reduced serum levels of thyroxin stem from lack of thyroxin secretion. They also suspected that PCB-induced liver enzymes hasten metabolism of thyroxin and contribute to low blood levels.

An early study (Kimura and Baba, 1973) demonstrated liver nodules in female but not male rats exposed to a total of 700-1500 mg Kanechlor 400 ingested over periods of time ranging from 22-80 weeks. Ito et al. (1974) exposed male Wistar rats to 0-1000 mg Kanechlor 300, Kanechlor 400 or Kanechlor 500/kg diet for 28-52 weeks. Nodular hyperplasia and cholangiofibrosis commonly occurred in high-level treatment groups but hepatocarcinomas were not found. Kimbrough et al. (1975) were able to produce hepatocellular carcinoma in female rats exposed to 100 mg Aroclor 1260/kg diet for 630 days. The only other finding of hepatocellular carcinoma in rats was reported by NCI (1977). Exposure to 50 or 100 mg Aroclor 1254/kg diet for 735 days was associated with hepatocarcinoma in 1/24 and 2/24, respectively; these incidences were not significant, but a significant dose-response relationship for liver nodules was demonstrated in this study. Ito et al.

(1973) demonstrated hepatocellular carcinoma in male mice exposed to 500 mg Kanechlor 500/kg diet for 224 days. Exposure to Kanechlor 400 or Kanechlor 300 at these levels and exposure to Kanechlor 500 at lower levels failed to produce liver cancer. Interestingly, mice did not exhibit the preneoplastic nodules that have been described in rats.

PCBs have been tested as a tumor-promoting agent in mice. Hepatocellular carcinoma was induced with α-BHC (Ito et al., 1973) in 8/30 mice exposed to 250 mg/kg diet for 224 days. The addition of 250 mg Kanechlor 500/kg diet for this period resulted in a doubling (15/26) of the incidence of hepatocellular carcinoma. Makiura et al. (1974) induced liver cancers in rats by administration of 3'-Me-DAB, 2-FAA or DEN in the drinking water for 24 weeks. Addition of 500 mg Kanechlor 500/kg to the diet seemed to have a protective effect and no cancers occurred in groups thus treated.

One report of gastric adenocarcinoma in rats was found. Morgan et al. (1981) investigated necropsy material from the NCI bioassay that exposed rats to levels of 0-100 mg Aroclor 1254/kg diet for 104-105 weeks. They found 6/144 gastric adenocarcinomas in treated rats and concluded that occurrence may have been tumor-related.

Aroclor 1254 appeared to be a weak tumor initiator (Di Giovanni et al., 1978) when tested in the two-stage mouse skin system with TPA as a promoting agent. Barry et al. (1978) also tested Aroclor 1254 as a tumor promotor in the two-stage mouse skin system initiated with DMBA but found no indication of promotion.

PCBs did not elicit positive response in most Ames Salmonella assays when tested with or without hepatic S-9 fraction from rats. One study seemed to be an exception. Wyndham et al. (1976) examined the mutagenicity of two pure PCBs α-isomers and two Aroclors. Although it was not clear from the text, it appeared that rabbit hepatic S-9 fractions was used for metabolic activator. They found pronounced mutagenicity of 4-monochlorobiphenyl but not more highly chlorinated species. Green et al. (1975) were unable to demonstrate dominant lethality in rats due to Aroclor 1254. Nilsson and Ramel (1974), using Drosophila melanogaster with sex-linked color markers, were unable to demonstrate clastogenic effects of Clophen 30 or Clophen 50. Cytogenic studies using Aroclor 1242 (Green et al., 1975a) or Aroclor 1254 (Green et al., 1975a; Garthoff et al., 1977; Dikshith et al., (1975) failed to demonstrate chromosomal aberrations in bone marrow or spermatogonia from PCB-treated rats.

Several workers investigated the teratogenicity and reproductive toxicity of various mixtures of PCBs. The only evidence of actual teratogenicity was found in mice. Marks et al. (1981) found an increased incidence of cleft palate, hydronephrosis and undersized renal papillae in fetal mice from dams that were exposed to 3,3',4,4',5,5'-HCB during days 6-15 of gestation. Watanabe and Sugahara (1981) also demonstrated increased incidence of cleft palate in fetal mice from dams that were exposed to Kanechlor 500. Both reports used oral dosage levels high enough to cause considerable maternal toxicity.

There are many reports of fetal toxicity. Linder et al. (1974) exposed rats to either Aroclor 1254 or 1260 for 1-2 generations. They did not report evidence of maternal toxicity, but found both reduced litter size and reduced survival at weaning. No terata were mentioned. Exposing pregnant rats to 30 mg Aroclor 1254/kg diet caused reduced litter weight and fetal death at delivery (Spencer, 1982). Fetal death, abortions and resorptions have been reported in rabbits (Villeneuve et al., 1971), rats (Baker et al., 1977) and mice (Orberg and Kihlstrom, 1973); again, terata were not mentioned. Truelove et al. (1982) produced fetal mortality and immunologic suppression in cynomolgus monkeys exposed to Aroclor 1254. One laboratory (Allen and Barsotti, 1976; Allen et al., 1979b; Allen et al., 1980) reported on the long-term exposure of rhesus monkeys to Aroclor 1248. Fetal death, abortions and underweight neonates were reported, and signs of neonatal toxicity included five infants facial and palpebral edema, hyperpigmentation and hair loss. These signs worsened after a nursing period of 4 months. Necropsy of infants that succumbed showed severe gastritis, lymphoid degeneration, hypocellular bone marrow and fatty liver but structural malformations were not found.

Mink appear to be especially sensitive to PCBs; as little as 5 mg Aroclor 1242/kg diet resulted in complete reproductive failure (Bleavins et al., 1980). Mice exposed to 0.025 mg Clophen A-60/day experienced lengthened estrus cycles (Orberg and Kihlstrom, 1973). Monkeys exposed to 2.5 mg Aroclor 1248/kg diet had irregular menstrual cycles with reduced levels of serum progesterone. Various workers have examined the effects of PCBs on male reproduction. Sager (1983) exposed neonatal male rats to PCBs from day 1 after birth by treating nursing dams with Aroclor 1254 by gavage. At 130 days of age these rats were observed for mating behavior. A significant reduction in the number of females impregnated by treated rats indicated interference with mating behavior. Testicular hyperplasia and subnormal development of accessory sex glands suggested that PCBs administered in the early postnatal period interfered with circulating levels of androgens and subsequent reproductive behavior.

VI. HEALTH EFFECTS IN HUMANS

Case Studies

Occupational.

Chloracne and Abnormal Hepatic Function -- Many case studies in the literature describe varying degrees of chloracne and abnormal hepatic function as a result of occupational exposure to PCBs. These studies were reviewed in detail by NIOSH (1977).

Early case studies of occupational PCB exposure were reported by Jones and Alden (1936), Drinker et al. (1937), Schwartz (1943) and Meigs et al. (1954). Jones and Alden (1936) reported 16 cases of chloracne among workers employed in the manufacture of PCBs; these are summarized in Table VI-1. These workers were also exposed to impure benzene. Patch tests on the workers with chloracne did not produce erythema or evidence of irritation.

Drinker et al. (1937) reported chloracne among workers (incidence not specified) breathing vapors containing 10% PCBs and 90% chlorinated naphthalenes. One of these workers also had what was described as jaundice.

Among four United States shipyards, 29 electricians were reported to have halowax acne (Schwartz, 1943). The electricians had worked with cables coated with halowax, which contains chlornaphthalenes and PCBs. The halowax acne developed after a few months of exposure and was most severe in cases of longer exposure.

TABLE VI-1

Summary of the Symptoms in Sixteen Cases of Acneform Eruption*

Case Number	Age (years)	Race	Duration of Exposure (months)	Type of Skin	Type of Eruption
1	22	white	6	seborrheic; previous acne	diffuse comedones; few cysts
2	28	white	5	average	diffuse comedones; few cysts
3	32	white	NR	seborrheic	diffuse comedones; large cysts and pustules
4	36	white	10	average dry	diffuse comedones; large cysts and abscesses
5	28	white	NR	average	diffuse comedones; few cysts on face and neck
6	30	white	8	seborrheic; previous acne	diffuse comedones; deep abscesses on neck; severe cysts
7	20	Negro	8	seborrheic	erythematous diffuse comedones; few small cysts
8	19	Negro	5	average	few scattered comedones; occasional cysts
9	26	Negro	NR	seborrheic	diffuse comedones; cysts; small abscesses
10	37	white	9	average	scattered comedones; occasional abscesses
11	56	white	NR	dry	scattered comedones; occasional abscesses

TABLE VI-1 (cont.)

Case Number	Age (years)	Race	Duration of Exposure (months)	Type of Skin	Type of Eruption
12	20	white	2	seborrheic	few comedones; occasional abscesses
13	37	white	NR	average	occasional comedones
14	23	Negro	12	average	very few comedones
15	22	white	NR	seborrheic	scattered comedones
16	20	Negro	NR	seborrheic	diffuse comedones; few cysts on back and face

*Source: Jones and Alden, 1936

NR = Not reported

Intermittent exposure to Aroclor vapors resulted in seven cases of chloracne among 14 workers 19 months after initiation of exposure (Meigs et al., 1954). The air concentration of Aroclor was measured 4 months before diagnosis of chloracne was made and this air concentration was determined to be 0.1 mg/m³. The mean length of exposure was 14.3 months for affected workers and 11.4 months for unaffected workers, but there was no good correlation between duration of exposure and workers with chloracne. The chloracne appeared to be a contact phenomenon, only developing on skin not covered by clothing. The seven workers affected by chloracne had normal blood cell counts, urinalysis and blood pressures. Of the seven workers with chloracne, six had normal liver function tests, which included direct and total bilirubin determinations and 24- and 48-hour cephalin flocculation, thymol turbidity and alkaline phosphatase determinations. The other affected worker had borderline cephalin flocculation and thymol turbidity, which was improved 13 months later.

Fischbein et al. (1979) reported a cross-sectional clinical field survey of 326 capacitor manufacturing workers (168 males, 158 females; mean years of employment, >15) exposed to various PCB mixtures (Aroclors 1254, 1242, 1016 and 1221). Both plants surveyed in this study also used chlorinated benzenes. The air levels (8-hour TWA) of PCBs in the plants were 0-11.0 mg/m³. Most workers had held different jobs within the plant, placing them in several different exposure levels. The duration of PCB exposure and age distribution by sex of the capacitor manufacturing workers are given in Tables VI-2 and VI-3, respectively. Dermatologic symptoms, including rash, burning sensation, acne, pigmentation (darkening), and thickening and discoloration of the fingernails, were reported by 76 of 168 males (45%) and by 87 of 168 females (55%). The prevalence of these dermatologic symptoms

TABLE VI-2

Duration of PCB Exposure of 326 Capacitor Manufacturing Workers*

Duration (years)	Number of Workers	Percent
<5	33	10.1
5.0-9.9	68	20.9
10.0-14.9	57	17.5
15.0-19.9	37	11.4
20.0-24.9	95	29.1
≥25.0	36	11.0

*Source: Fischbein et al., 1979

TABLE VI-3

Age Distribution of 326 Capacitor Manufacturing Workers by Sex*

Age (years)	Total Number Examined	Males	Females
<30	49	33	16
30-39	61	41	20
40-49	88	48	40
50-59	93	27	66
60-69	25	13	12
≥70	10	6	4
Total	326	168	158

*Based on data from Fischbein et al., 1979 is given in Table VI-4. Warshaw et al. (1979) reported the pulmonary and ocular symptoms given by these workers to be cough (13.8%), wheezing (3.4%), tightness in chest (10.1%) and upper respiratory or eye irritation (48.2%). Fischbein et al. (1979) reported the gastrointestinal, musculoskeletal and neurologic complaints of these workers, as summarized in Table VI-5. Gastrointestinal symptoms were reported by 58 of 326 workers (18%; 22 males, 36 females). Neurologic symptoms were reported by 64 of 168 males (39%) and 94 of 168 females (58%), but no remarkable prevalence of any neurologic effect was observed during physical examinations.

Fischbein et al. (1979) performed physical examinations on the 326 PCB-exposed capacitor manufacturing workers and found abnormal dermatologic effects, including erythema, swelling, dryness and thickening, in 68 of 168 males (41%) and 60 of 168 females (38%). In addition, 16 of 326 (5%) had acneform eruptions; 48 of 326 (15%) had conjunctival and palpebral abnormalities, including palpebral hyperpigmentation and edema; 7 of 326 (2%) had abnormal eye secretions and 3 of 326 (1%) had enlarged meibomian glands. There were only a few scattered abnormal results found in biochemical and hematologic tests. In pulmonary function studies, a decreased forced vital capacity was seen in 34 of 243 workers examined (14%), but no abnormalities were seen in chest X-rays. Overall, there was no correlation between any symptoms and duration of employment. There was, however, a significant association between plasma PCB level and increased SGOT levels.

Maroni et al. (1981) reported blood PCB concentrations of 41-1319 ppb in 80 electrical workers (40 male, 40 female) employed in electric capacitor manufacture and testing plants. Of the 80 workers, 67 were exposed to

TABLE VI-4

Prevalence of Reported Dermatologic Symptoms Among 326 Capacitor Manufacturing Workers*

Symptom	Number	Percent
Rash	128	39.3
Burning sensation	81	24.8
Acne	35	10.7
Pigmentation (darkening)	8	2.5
Thickening	12	3.7
Discoloration of fingernails	8	2.5

*Source: Fischbein et al., 1979

TABLE VI-5

Prevalence of Gastrointestinal, Musculoskeletal and Neurologic Symptoms by Symptom Category and Sex Among 326 Capacitor Manufacturing Workers*

Symptom	Males		Females	
	Number	Percent of Group Examined	Number	Percent of Group Examined
Gastrointestinal:				
Anorexia	8	4.8	3	1.9
Weight loss	16	9.5	8	5.1
Nausea	7	4.2	14	8.9
Vomiting	4	2.4	2	1.3
Abdominal pain	5	3.0	15	9.5
Musculoskeletal:				
Muscle pain	5	3.0	6	3.8
Joint pain	13	7.7	24	15.2
Neurologic:				
Headache	33	19.6	44	27.8
Dizziness	16	9.5	22	13.9
Depression	5	3.0	19	12.0
Memory loss	2	1.2	13	8.2
Fatigue	17	10.1	36	22.8
Nervousness	21	12.5	49	31.0
Sleeplessness	10	6.0	17	10.8
Somnolence	8	4.8	18	11.4

*Based on data from Fischbein et al., 1979

Pyralene 3010 (a PCB mixture containing 42% chlorine) and 13 were exposed to Apirolio (a PCB mixture containing 42% chlorine). The mean age of the workers was 37±8 years, and the mean duration of employment was 12±6 years. There were six cases of chloracne among the 80 workers, as shown in Table VI-6. Sixteen of the males had liver abnormalities, including hepatomegaly and hepatic dysfunction (indicated by an increase in serum enzymatic activities); for 20% of these, the blood PCB concentration was <200 ppb. No liver abnormalities were reported among exposed female workers. There were two cases of bleeding hemangioma, one of whom also had chronic myelocytic leukemia.

Guw et al. (1976) compared dermatologic and hepatic parameters of 34 electrical industry workers exposed to electrical grade (no impurities) Aroclor 1242 with those of 30 control volunteers. The electrical workers consisted of 15 males (6 months to 23 years employment) and 19 females (1 month to 7 years employment), and were identified by two cohorts, impregnation room workers (14 males, 5 females) and process workers (1 male, 14 females). Potential for PCB exposure was much higher for the impregnation room workers than for the process workers. The control volunteers had no history of occupational exposure to PCBs and had no measureable blood levels of PCBs. Hepatic function tests on both cohorts of workers yielded scattered abnormal values, but when considering both groups as a whole, the mean value for each test is within normal limits. Major worker complaints consisted of burning of the eyes, face and skin. One worker had chloracne without systemic effects, and five workers had eczematous hand and leg rashes. These dermatologic effects occurred at an air concentration of PCBs of <1 mg/m³. There was no clear correlation between the level of Aroclor

TABLE VI-6

Clinical Features of Six Electrical Workers with Chloracne*

			Skin Lesions		
Case No.	Age (years)	Age at First PCB Exposure	Age at Onset	Current Findings	Affected Regions
1	49	27	39	vermicular scars, comedones, superficial cysts and suppurative folliculitis	abdomen, thighs
2	26	16	22	vermicular scars, comedones, superficial cysts, and suppurative folliculitis	face, neck
3	50	20	23	vermicular scars, comedones, superficial cysts, and supperative folliculitis	neck, shoulders arms, back
4	43	28	40	vermicular scars	arms, back, legs
5	38	24	37	folliculitis (possibly chloracne)	scrotum
6	49	42	45	comedones with erythema (possibly chloracne)	neck, sternum

*Source: Maroni et al., 1981

exposure, dermatitis severity, and complaints of skin burning or irritation; however, complaints were recorded more frequently for those workers having higher blood levels of Aroclor. In contrast to the findings of Maroni et al. (1981), there were no significant health effects in workers at or below a blood PCB level of 200 ppb.

PCBs have been reported to alter drug metabolism through the mechanism of hepatic enzyme induction in exposed workers (Alvares et al., 1977). A significantly decreased mean antipyrine half-life (10.8 hours) in five exposed workers as compared with nonexposed control subjects (half-life of 15.6 hours) was reported. No systemic toxic effects were reported among the PCB-exposed workers.

Zauder (1979) associated hepatocellular damage in a woman with concomitant occupational exposure to PCBs and medical exposure to halothane anesthesia. Zauder (1979) further speculated that there was a correlation between this human case and the animal model of halothane hepatic damage elucidated by Sipes and Brown (1976) (Chapter VII).

Malignant Melanoma -- Bahn et al. (1976, 1977) reported an increased incidence of malignant melanomas in employees of a Northeastern United States petrochemical plant that had used Aroclor 1254 for 9 years (ending in the late 1950s). Of 31 research and development employees subject to "heavy" Aroclor exposure (quantified exposure levels not reported), two (0.065%) had malignant melanomas. This incidence was significantly increased (p=0.001); based on a person-year analysis and the standard of the

Third National Cancer Survey incidence rates in which only 0.04 malignant melanomas would be expected among 31 persons (NCI, 1975). Of a group of 41 refinery workers exposed to "low" levels of Aroclor (quantified exposure levels not reported), one had malignant melanoma. Exposure to other potential or known carcinogenic substances was not defined. The NIOSH report (1977) further examined the data from this preliminary study. This report indicates that there were a total of 8 cancers which included 3 melanomas and 2 pancreatic cancers. These two types of malignancies were present in higher than expected incidences. Final data were not available.

Clinical Case Studies. High PCB serum levels were found in some women who had recent or former missed abortions (mean PCB serum levels of 103.04, 82.00 and 20.69 ppb for recent missed abortions, former missed abortions and control groups, respectively) (Bercovici et al., 1983) and in some women with premature delivery (mean PCB serum levels of 128 ppb in the premature delivery group vs. 26.5 ppb in the control group) (Wassermann et al., 1982). Bercovici et al. (1983) defined incomplete or missed abortion as "the retention in the uterus of the products of conception 6 weeks or more after the death of the fetus that occurred before the 20th completed week of gestation." The higher PCB serum levels were associated with increased incomplete abortions (Bercovici et al., 1983) and premature deliveries (Wassermann et al., 1982), but a definitive causal relationship cannot be established, as only small numbers of women were examined (up to 17 symptomatic; up to 10 asymptomatic), and some of these women had high serum levels of some organochlorine insecticides (DDT isomers and their metabolites, lindane, dieldrin, heptachlor epoxide).

Epidemiological Studies

Yusho Incident. The best documentation of human effects as a result of ingestion of PCBs can be derived from the Japanese poisoning incident that occurred in 1968. The more than 1000 persons affected by Yusho, or oil disease, had ingested rice oil contaminated with a commercial brand of PCBs, Kanechlor-400, used as a heat transfer agent in the manufacture of the oil. The average total amount of PCBs consumed was estimated to be ~2 g, with ~0.5 g being the least total amount consumed by an affected group of 325 people (Kuratsune et al., 1972). The contaminated oil also contained ~5 ppm of polychlorinated dibenzofurans (PCDFs), some 250 times more than the concentration generally found in Kanechlor-400 that has not been reheated for long periods of time (Nagayama et al., 1976). The presence of the potent toxicants PCDFs in the Yusho oil may have contributed heavily to the overall toxicologic effects seen in Yusho patients (Nagayama et al., 1976).

Kuratsune et al. (1972) did an epidemiologic study on 325 (158 male, 167 female) Yusho patients. The most common initial symptoms were increased eye discharge and swelling of upper eyelids (38.3%) followed by acneform eruptions and follicular accentuation (33.1%) and pigmentation of the skin (9.6%) (Goto and Higuchi, 1969). These inital Yusho symptoms, reported among 136 patients, are summarized in Table VI-7.

Kuratsune et al. (1969) reported that the most notable symptoms of Yusho among 189 patients included dark brown pigmentation of nails and skin, follicular accentuation, acneform eruptions, increased eye discharge, increased sweating at the palms and feeling of weakness. The percent distribution of symptoms among 189 Yusho patients is summarized in Table VI-8.

TABLE VI-7

Initial Symptoms of Yusho Among 136 Persons*

Initial Symptoms	Patients	
	No.	%
Swelling of upper eyelids, increased eye discharge	52	38.3
Acne-form eruption, follicular accentuation	45	33.1
Edematous swelling of limbs	9	6.6
Languishment	4	2.9
Disturbances in digestive canal	4	2.9
Numbness and other neurological signs	9	6.6
Pigmentation of skin	13	9.6
Total	136	100.0

*Source: Goto and Higuchi, 1969

TABLE VI-8

Percent Distribution of Symptoms of Yusho Among 189 Persons*

Symptoms	Males (n=89)	Females (n=100)
Dark brown pigmentation of nails	83.1	75.0
Distinctive hair follicles	64.0	56.0
Increased sweating at palms	50.6	55.0
Acnelike skin eruptions	87.6	82.0
Red plaques on limbs	20.2	16.0
Itching	42.7	52.0
Pigmentation of skin	75.3	72.0
Swelling of limbs	20.2	41.0
Stiffened soles in feet and palms of hands	24.7	29.0
Pigmented mucous membrane	56.2	47.0
Increased eye discharge	88.8	83.0
Hyperemia of conjunctiva	70.8	71.0
Transient visual disturbance	56.2	55.0
Jaundice	11.2	11.0
Swelling of upper eyelids	71.9	74.0
Feeling of weakness	58.4	52.0
Numbness in limbs	32.6	39.0
Fever	16.9	19.0
Hearing difficulties	18.0	19.0
Spasm of limbs	7.9	8.0
Headache	30.3	39.0
Vomiting	23.6	28.0
Diarrhea	19.1	17.0

*Source: Karatsune et al., 1969

Based on the estimated amounts of Kanechlor-contaminated rice oil consumed in Table VI-9 and the clinical severity of resulting effects of different age groups in Table VI-10, Kuratsune et al. (1972) demonstrated a qualitative dose-response relationship.

Hirayama et al. (1974) reported an inverse correlation between serum bilirubin concentration in Yusho patients and blood PCB levels, with mean serum bilirubin concentrations of 0.48±0.26 mg/100 mℓ in 121 Yusho patients and 0.87±0.33 mg/100 mℓ in 257 healthy adult controls. Increased serum triglyceride concentration was also seen in Yusho patients.

Shigematsu et al. (1978) reported respiratory effects of expectoration (40%), mild wheezing (2%), bronchiolitis (many) and pneumonia or atelectasis (~10%) among 289 nonsmoking Yusho patients.

Yamashita (1977) reported four cases of infants born to mothers who had Yusho during prenancy. The amount of PCB contaminated oil consumed during pregnancy was ~1.1-10.5 ℓ. Maternal symptoms included acneform eruptions, follicular accentuation; dark brown pigmentation on the skin, mucous membranes and nails; and hypersecretion of the Meibomian gland. Three of the four infants, including one full-term (40 weeks gestation), one premature (36 weeks gestation), and one 2 weeks later than term (42 weeks gestation), were small-for-gestational age (both weight and height). Other clinical features among the four infants included dark brown pigmentation on the skin and mucous membranes, gingival hyperplasia, eruption of teeth at birth, spotted calcification on the parieto-occipital skull and the large or wide fontanels and sagittal suture, facial edema and exophthalmic eyes.

TABLE VI-9

Relationship Between the Amount of Kanechlor-Contaminated Rice Oil Consumed and Clinical Severity of Yusho*

Estimated Amount of Oil Consumed	Nonaffected		Light Cases		Severe Cases		Total	
	No.	%	No.	%	No.	%	No.	%
<720 mℓ	10	12	39	49	31	39	80	100
720-1440 mℓ	0	0	14	31	31	69	45	100
>1440 mℓ	0	0	3	14	18	86	21	100

*Source: Kuratsune et al., 1972

TABLE VI-10

Relationship Between Clinical Severity of Yusho and Age of Affected Persons[a]

Estimated Amount of Oil Consumed	Age (years)	Number of Patients By Clinical Severity							
		Nonaffected		"Light"[b]		"Severe"[b]		Total	
		No.	%	No.	%	No.	%	No.	%
<720 mℓ	<12	3	16.7	13	72.2	2	11.1	18	100
	13-29	2	8.3	7	29.2	15	62.5	24	100
	≥30	5	13.1	19	50.0	14	36.9	38	100
	Total	10	12.5	39	48.8	31	38.7	80	100
720-1440 mℓ	<12	0	0	5	50.0	5	50.0	10	100
	13-29	0	0	1	6.7	14	93.3	15	100
	≥30	0	0	8	40.0	12	60.0	20	100
	Total	0	0	14	31.2	31	68.8	45	100

[a] Source: Kuratsune et al., 1972

[b] Clinical severity was classified as "light" or "severe," as defined by Goto and Higuchi (1969); further details enumerating clinical features of this classification scheme were not reported.

Kuratsune et al. (1969) summarized four studies (Yamaguchi et al., 1971; Taki et al., 1969; Funatsu et al., 1971; Kikuchi et al., 1969) of 10 live and 3 stillborn births from February 15 to January 31, 1968, to 11 females with Yusho during pregnancy and 2 wives of males with Yusho during the female's pregnancy. The amount of Kanechlor-contaminated oil consumed during pregnancy was 0.3-2.6 ℓ (Yamaguchi et al., 1971). Of 10 live and 2 stillborn births, 9 had unusually grayish, dark-brown stained skin, 5 had similar pigmentation of the gingiva and nails and most had increased eye discharge (Yamaguchi et al., 1971; Taki et al., 1969; Funatsu et al., 1971). The third stillborn infant had marked epidermal hyperkeratosis and atrophy; cystic dilation of hair follicles, especially on the head; and a marked increase of melanin pigments in epidermal basal cells (Kikuchi et al., 1969). Of the 13 infants, 12 were described as smaller than the national Japanese standards and 4 as small-for-dates babies (Yamaguchi et al., 1971; Taki et al., 1969); however, small was not defined in terms of height, weight or any other clear measurable difference.

Yoshimura (1971) compared the growth of 42 school-aged children (23 males, 19 females) with Yusho with that of 719 sex-matched classmates described as being "healthy". For the years 1967-1969, the height and weight gains of girls with Yusho were not affected, while boys with Yusho had significantly less height and weight gains.

Carcinogenicity Data from Yusho Incident -- By 1979, 31 Yusho patients had died, 11 (35.4%) of these from malignant neoplasms (Urabe et al., 1979).

Only 21.1% of all deaths in this Japanese prefecture would be expected from malignant neoplasms, but no clear correlation between the occurrence of Yusho and increased deaths from malignant neoplams could be made because of the small number of deaths observed and unknown latent period. The 11 malignant neoplasms included 2 stomach cancers, 1 stomach and liver cancer (possibility of metastasis not discussed), 2 liver cancers with cirrhosis, 2 lung cancers, 1 lung tumor, 1 breast tumor and 2 malignant lymphomas. No analyses were made on the significance of the actual vs. the expected incidence of a particular tumor type. The Yusho incident data, therefore, do not give a clear picture of whether or not there was an increase in cancer among the affected population.

Yu-Cheng Incident. In the spring of 1979, ~2000 persons in the central western part of Taiwan presented symptoms similar to those seen with Yusho in Japan; these symptoms were collectively referred to as Yu-Cheng (Chang et al., 1980a,b, 1981; Chen et al., 1980, 1981). The affected persons had consumed rice-bran oil contaminated with PCBs that were used as a heat transfer medium in the manufacture of the oil. Blood PCB levels of 66 affected persons ranged from 11-720 ppb (mean 49 ppb; 39 ppb if the patient with the blood level of 720 ppb is excluded) at ~9-12 months after consumption of the PCB-contaminated oil (Chen et al., 1980). These blood PCB levels were much higher than those reported for the Japanese Yusho incident, probably because of the larger percent of higher chlorinated PCBs in the Chinese oil and the smaller elapsed time period between consumption of the contaminated-oil and blood PCB level measurements (Chen et al., 1980, 1981). The concentration of PCBs and PCDFs in six different samples of the contaminated oil are given in Table VI-11.

TABLE VI-11

Concentration of PCBs and PCDFs in the Toxic Rice-Bran Oils That Caused "PCB Poisoning" in Taichung, Taiwan*

Sample No.	PCB (ppm)	PCDF (ppm)	PCB/PCDF Ratio
1	405	1.68	241
2	53	0.180	294
3	99	0.399	248
4	78	0.250	312
5	77	0.209	368
6	65	0.297	219

*Source: Chen et al., 1981

The most common symptoms among persons consuming the PCB-contaminated oil were acneform eruptions and follicular accentuation, skin and nail pigmentation, swelling of the eyelids and increased discharge from the eyes, headache, nausea and numbness of the limbs (Chang et al., 1980b). The major blood disorders were decreased erythrocyte counts, increased white blood cell counts, decreased concentrations of hemoglobin and gamma-immunoglobulin, increased serum triglyceride and increased SGOT, SCPT and serum alkaline phosphate activities (Chang et al., 1980a). Decreased concentrations of γ-aminolevulinic acid and uroporphyrin were also seen in affected persons (Chang et al., 1980b).

Effects on Blood Pressure Measurements. Kreiss et al. (1981) examined 458 volunteers from Triana, Alabama, ≥12 years of age, and correlated serum PCB levels (Aroclor 1260) with elevated blood pressure. The mean serum PCB level among this group was 17.2 ppb. Three classifications of blood pressure measurements were used: normal (systolic of <140 mm Hg and diastolic of <90 mm Hg), borderline hypertension (systolic of 140-159 mm Hg and diastolic of 90-94 mm Hg) and hypertension (systolic of ≥160 mm Hg and diastolic of ≥95 mm Hg). The incidence of borderline and definite hypertension was increased 30% more than would be expected for a general population wth the same age, race and sex composition.

Ocular Melanoma. Davidorf and Knupp (1979) conducted an epidemiological study of ocular melanoma incidence in Ohio from 1967-1977, attempting to associate Ohio counties with known high concentrations of PCBs and those with industries that might use PCBs with an increased incidence of ocular melanoma as compared with the statewide incidence. In determining the tumor incidence for ocular melanoma, non-whites and persons under 21 years old were excluded, as white persons over 21 years old are considered to be the susceptible population. There were 698 cases (average age, 58.1 years) of primary choroidal melanomas in Ohio during the 11-year period 1967-1977, corresponding to a statewide tumor incidence of 1.09 cases/100,000 persons. About half of these 698 cases were found in tumor registries, and the rest were identified from individual medical records, ophthalmologists and ocular pathology laboratories. When compared with the total 11-year statewide incidence (1.09 cases/100,000 persons), there was no statisically significant increase in the yearly ocular melanoma tumor incidence for any individual Ohio county over the 11-year period. The incidence of ocular melanoma was increased, but not significantly, for one county (Jackson County); however, no correlation could be made for the existing distribution of ocular melanoma, PCBs or industries that might use PCBs. Davidorf and Knupp (1979) concluded that their results did not suggest a causal relationship between PCB exposure and an increased annual occurrence of ocular melanoma in Ohio counties for the 11-year period of 1967-1977. Davidorf and Knupp (1979) noted, however, that some areas were under-represented in the study, and also that the inhabitants of these poor rural counties were less likely to seek early medical care.

High Risk Subpopulations

Because of the potential for frequent or high exposure, persons exposed to PCBs occupationally are at high risk. Another group of high risk because of the potential for high exposure is nursing infants, as PCBs are excreted in the breast milk of lactating women (see Distribution Section in Chapter III) (Miller, 1977; Rogan et al., 1980; Wolff, 1983; Wickizer et al., 1981; Mes and Davies, 1979; Kuwabara et al., 1979; Hofrander et al., 1981).

Evidence of human teratogenic and embryotoxic effects from maternal PCB exposure can be seen in the infants of women who had Yusho during pregnancy (see Yusho Incident Section) (Yamashita, 1977). This identifies both the pregnant human and the fetus as high risk subpopulations. Yamashita (1977) reported that children born to women who had Yusho during pregnancy were small for gestational age, and had dark brown pigmentation of the skin and mucous membranes, gingival hyperplasia, eruption of teeth at birth, spotted calcification on the parieto-occipital skull and the large or wide fontanels, sagittal suture, facial edema and exophthalmic eyes.

Embryos, fetuses and neonates (2-3 months old) are a subpopulation at special risk because of inherent physiological differences from the adult human. This subpopulation usually lacks the hepatic microsomal enzyme systems, including the glucuronidation pathway, that are capable of oxidizing PCBs to facilitate the detoxification and excretion of these compounds (Calabrese and Sorenson, 1977; Gillette, 1967; Nyhan, 1961). Breast-fed infants are at greater risk also because of a steroid excreted in human breast milk, but not in cow's milk, that inhibits glucuronyl transferase activity, and thus glucuronidation and excretion of toxicants such as PCBs, by >20% (Calabrese and Sorenson, 1977; Gartner and Arias, 1966). Children simultaneously exposed to the antibiotic novobiocin and PCBs may be unable to effectively detoxify and excrete PCBs, as novobiocin noncompetitively inhibits glucuronyl transferase activity _in vitro_ (Lokietz et al., 1963; Calabrese and Sorenson, 1977).

Persons with improperly or incompletely developed glucuronide conjugation systems, such as those with Gilbert's syndrome or Crigler and Najjar syndrome, may be subpopulations at high risk because of the potential ineffectiveness to detoxify and excrete PCBs (Lester and Schmid, 1964; Calabrese and Sorenson, 1977). Also, persons with hepatic infections (i.e., infectious hepatitis) may have depressed glucuronide synthesis, making them a subpopulation at high risk (Calabrese and Sorenson, 1977).

Summary

Many studies of occupational exposure to PCBs have shown varying degrees of dermatologic and hepatic effects (Jones and Alden, 1936; Drinker et al., 1937; Schwartz, 1943; Meigs et al., 1954; Fishbein et al., 1979; Warshaw et al., 1979; Maroni et al., 1981; Ouw et al., 1976; Alvares et al., 1977; Zauder, 1979). These effects were produced at airborne PCB levels as low as 0.1 mg/m^3 (Meigs et al., 1954), and Alvares et al. (1977) reported impaired liver function in the absence of overt toxic effects. Bahn et al. (1976, 1977) reported a significantly increased incidence of malignant melanoma in petrochemical workers exposed to PCBs and other unidentified chemicals.

High PCB serum levels among pregnant women have been associated with increased missed abortions (Bercovici et al., 1983) and premature deliveries (Wassermann et al., 1982).

Accidental human ingestion of PCB-contaminated rice oil in Japan and Taiwan resulted in effects silar to those seen following occupational exposure (e.g., acneform eruptions, abnormal hepatic function) (Kuratsune et al., 1969, 1972; Goto and Higuchi, 1969; Hirayama et al., 1974; Chang et al., 1980a,b; Chen et al., 1980, 1981). Infants born to Japanese women who had Yusho during pregnancy were generally small-for-gestational age and had dark brown pigmentation on the skin and mucous membranes, gingival hyperplasia, eruption of teeth at birth, spotted calcification on the parieto-occipital skull and the large or wide fontanels and sagittal suture, facial edema and exophthalmic eyes (Yamashita, 1977; Yamaguchi et al., 1971; Taki et al., 1969; Funatsu et al., 1971; Kikuchi et al., 1969).

Kreiss et al. (1981) found a direct correlation between increasing serum PCB levels and elevated blood pressure in 458 volunteers ($\geq$12 years of age) from Triana, Alabama. Davidorf and Knupp (1979) did not find a correlation between the incidence of ocular melanoma in Ohio and the distribution of PCBs either environmentally or industrially.

High risk subpopulations include those with the potential for frequent or high exposure, including occupationally-exposed workers and nursing infants, as PCBs are excreted in the breast milk of lactating humans (Miller, 1977; Rogan et al., 1980; Wickizer et al., 1981; Mes and Davies, 1979; Kuwabara et al., 1979; Hofrander et al., 1981). Because of the birth defects seen in infants of Yusho patients (Yamashita, 1977), fetuses and pregnant women are clearly high risk subpopulations. Other high risk subpopulations are based on the inability to oxidize PCBs by glucuronidation, decreasing their capability to detoxify and excrete these xenobiotic compounds; these high risk subpopulations include, but are not limited to embryos, fetuses, neonates (2-3 months old), especially breast-fed infants, children simultaneously exposed to the antibiotic novobiocin, persons with Gilbert's syndrome or Crigler and Najjar syndrome and persons with hepatic infections, such as infectious hepatitis (Calabrese and Sorenson, 1977; Gillette, 1967; Nyhan, 1961; Gartner and Arias, 1966; Lokeitz et al., 1963; Lester and Schmid, 1964).

VII. MECHANISMS OF TOXICITY

Enzyme Induction

PCBs have been demonstrated to be potent enzyme inducers in a wide variety of species including rats, rabbits, ferrets, monkeys, mink and humans (Risebrough et al., 1968; Street et al., 1969; Wolff and Hesse, 1977; Lake et al., 1979; Iverson et al., 1982; Shull et al., 1982). A number of authors have investigated the relationship between the degree of chlorination and induction potency. Litterst et al. (1972) fed diets containing 0.5, 5.0, 50 or 500 ppm (0.5, 5.0, 50 or 500 mg/kg diet) of Aroclors 1242, 1248, 1254 or 1260 to groups of 6 male Osborne-Mendel rats for 4 weeks. They found that the hepatic enzyme induction observed increased with increasing chlorine content. Similar results have been reported in Sprague-Dawley rats treated with Aroclor 1016 or 1254 (Bickers et al., 1972). Since a positive correlation has been observed between the long-term toxicity and degree of chlorination of commercial PCB mixtures (see Health Effects in Animals in Chapter V), the ability of PCBs to induce enzymes may be related to their toxicities.

Two general types of enzyme inducers are recognized, typified by 3-methylcholanthrene (P-448 inducers) and phenobarbital (P-450 inducers). Alvares et al. (1973) found that Aroclor 1254 produced both types of induction in male Sprague-Dawley rats, and concluded that PCBs represented a third, more general, type of inducer. This situation was clarified by Goldstein et al. (1977) who measured the enzyme induction produced by various isomerically pure PCBs. They found that substitution at both para positions was necessary for potent enzyme induction. In addition, those biphenyls that were chlorinated symmetrically at both the meta and para positions were P-448 inducers, while chlorination at the ortho and para, or ortho, meta and para positions resulted in the induction of P-450. Goldstein et al. (1977) also noted that the isomers that induce P-448 were also the most toxic PCBs, as measured by weight loss.

Yoshimura et al. (1979) investigated the relationship between P-450 or P-448 induction and the toxicity of several individual PCBs. They found that those PCBs which induced P-450 (phenobarbital-type inducers) were not significantly toxic to male Wistar rats at a dose of 100 mg/kg bw, while those that induced P-448 (3-methylcholanthrene-type) reduced growth rate, reduced thymus and spleen weights and produced liver enlargement accompanied by fatty liver at a dose of 10 mg/kg bw. The extent of the toxic response was correlated with the potency of P-448 induction; however, 3-methylcholanthrene treatment did not produce similar signs of toxicity. This suggests that there is no causal relationship between induction of P-448 and toxicity.

Shull et al. (1982) compared the toxicities and enzyme induction potencies of Aroclor 1242 and 1016 in mink and ferrets. Aroclor 1242 was the more potent toxicant and inducer in both species. There was, however, an inverse relationship in species sensitivity to the enzyme inductive effects and toxicity of these mixtures. It, thus, appears that relative P-448 enzyme induction may be correlated with toxicity within a given species, but not between species.

Inhibition of Mitochondrial Respiration

Sivalingan et al. (1973) investigated the effect of KC-300, KC-400 and KC-500 (mixtures of tri-, tetra- and pentachlorobiphenyl isomers, respectively) on isolated rat liver mitochondria. KC-300 was an inhibitor of the electron transfer system at all concentrations tested (10^{-2} to 10^{-9}M). KC-400 and KC-500 inhibited the electron transport system at low concentrations, but produced an uncoupling effect at higher concentrations. In contrast, Hasegawa et al. (1982) found tetrachlorinated biphenyls to be inhibitors of respiration in liver mitochondria isolated from male Donryu rats at concentrations of up to 100 μM.

Khan and Cutkomp (1982) determined the effect of Aroclor 1242 and 1254 on mitochondria isolated from the red coxal muscle of adult male cockroaches. At concentrations of 6-12 μM or above, Aroclor 1254 and 1242 produced an uncoupling of respiration. The results of these and other investigators (Pardini, 1971; Desaiah et al., 1972; Cutkomp et al., 1972; Nishihara, 1983; Koch et al., 1972) indicate that the effects of PCBs on mitochondrial respiration varies with the mitochondrial source, the concentration of PCB and the degree or pattern of chlorination.

Reproductive Toxicity

Bitman and Cecil (1970) investigated the estrogenic effects of a number of chlorinated hydrocarbons structurally related to DDT. Estrogenic activity was measured by the 18-hour glycogen response of the uterus of immature Wistar rats. PCBs tested included Aroclor 1221, 1232, 1242, 1248, 1254, 1260, 1262, 1268 and 4465. They found that all the PCBs with <54% chlorination, below Aroclor 1254, induced an estrogenic response. Similar results were reported by Gellert (1978) who found that Aroclor 1221, but not Aroclors 1242, 1254 or 1260, would induce a uterotrophic response in weanling Sprague-Dawley rats.

Fuller et al. (1980) investigated the effect of in vivo exposure to Clophen A-30 (20 mg/kg bw, 30% chlorine) on in vitro progesterone synthesis in tissue from serum-human chorionic gonadotropin primed immature rats (strain unspecified). PCB treatment resulted in an ~100% increase in progesterone synthesis above control levels.

PCBs also alter endocrine function by inducing hepatic enzymes responsible for steroid metabolism. Orberg and Ingvast (1977) investigated the effect of 2,4'5-trichlorobiphenyl or 2,2',4,4',5,5'-hexachloro-biphenyl on the plasma clearance of $[4\text{-}^{14}C]$ progesterone derived radioactivity in female Sprague-Dawley rats. When the rats were treated with 0.5 mg/day for 14 consecutive days, 2,2',4,4',5,5'-hexachlorobiphenyl, but not 2,4',5-trichlorobiphenyl, significantly increased the elimination rate of ^{14}C from plasma. Derr (1978) demonstrated that in vivo homeostatic mechanisms were insufficient for the maintenance of normal progesterone levels in female Sprague-Dawley rats dosed with 10 or 40 mg Aroclor 1254 in sesame oil/kg bw every third day for 2 weeks. PCBs that induce hepatic enzymes (see Enzyme Induction) also increase the rate of metabolism and lower plasma levels of estradiol-17β and testosterone (Derr and Dekker, 1979; Conney et al., 1967; Kintzman et al., 1967).

Thus, PCBs may both have a direct estrogenic effect and alter the metabolism and plasma levels of steroid hormones. Both of these mechanisms may be involved in the reproductive toxicity of PCBs.

Mobilization and Redistribution of Stored PCBs

PCBs are highly lipophilic compounds that are stored in body fat for long periods of time (see Bioaccumulation and Retentions in Chapter III).

During period of stress, these fat deposits may be mobilized, releasing the stored PCBs into the systemic circulation where they may exert their toxic effects. Carlson (1980) administered oral doses of 0 or 10 mg Aroclor 1254 in corn oil/kg bw/day to groups of five Sprague-Dawley rats for 14 days. Animals were sacrificed from 1-148 days following the last dose and hepatic enzyme activities were measured. Enzyme induction persisted for 4-5 months. This induction was enhanced if the animals were starved for 4 days, thus presumably mobilizing stored PCBs, before measurement of enzyme activities.

Covalent Binding

Wyndham and Safe (1978) investigated the *in vitro* microsomal mediated binding of 4-chlorobiphenyl and 4'-chloro-4-biphenylol to protein and nucleic acids. Uninduced microsomes mediated binding between the chlorinated biphenyls and microsomal protein, but not exogenous RNA or DNA. When microsomes from Aroclor 1248 pretreated rats were used, binding to microsomal protein was enhanced and measurable binding to endogenous and exogenous nucleic acids was observed. Binding levels were too low for the determination of the specific RNA or DNA adducts formed.

Shimada and Sato (1980) investigated the *in vitro*, microsomal mediated binding of ^{14}C-labeled KC-300, KC-500, 2,4,2',5'-tetrachlorobiphenyl, 2,4,2',4'-tetrachlorobiphenyl and 2,4,5,2',4',5'-hexachlorobiphenyl to endogenous microsomal protein. Binding was enhanced by pretreatments with inducers of the microsomal mono-oxygenase system and by 1,1,1-trichloropropene-2,3-oxide, an inhibitor of epoxide hydrase. No binding was observed with 2,4,5,2',4',5'-hexachlorobiphenyl, which does not have two adjacent unchlorinated carbon atoms. These data suggest that the macromolecular binding of PCBs is mediated by the metabolic formation of PCB epoxides. This macromolecular binding may be the mechanism by which PCBs induce their mutagenic and carcinogenic effects (see Carcinogenicity and Mutagenicity in Chapter V).

Altered Calcium Metabolism

Accumulation of calcium in the heart and kidneys has been observed in individuals chronically poisoned with PCB-contaminated rice oil (Kikuchi and Masuda, 1973; see Case Studies in Chapter VI), indicating that PCBs may alter calcium metabolism. Yagi et al. (1976) fed diets containing 0 or 500 ppm (0 or 500 mg/kg diet) KC-500 to groups of 10 male Wistar rats for 30 days. At autopsy, sodium and magnesium levels were decreased in the blood, liver and kidneys, potassium levels were decreased in the heart and kidneys, and calcium levels were decreased in the bone, spinal cord and sciatic nerve and increased in the kidneys.

Immunological Effects

Suppression of the immune system is one of the more sensitive indicators of PCB intoxication (see Immunological Effects in Chapter V). Conradt et al. (1979) suggested that PCBs may suppress immune function by inhibiting RNA synthesis in macrophages. They found that Aroclor 1242 (>7 μg/mℓ) inhibited *in vitro* RNA synthesis, as measured by [^{3}H] uridine incorporation, in resting macrophages. Since RNA synthesis is suspected to be involved in the acquisition and transfer of immunity (Gottlieb and Waldman, 1972), inhibition of macrophage RNA synthesis may represent a mechanism by which PCBs induce immunosuppression. Suppression of the immune system may, in turn, produce other signs of toxicity because of decreased resistance to infection or tumor formation.

Thyroid Toxicity

PCBs have been observed to induce numerous alterations in the structure and function of the thyroid gland (see Thyroid Effects in Chapter V). In the normally functioning thyroid, there is a continuous turnover of iodine as the iodine is actively accumulated and converted to thyroglobulin, the thyroglobulin is proteolyzed, and thyroxine and tri-iodothyronine are secreted. Iodine accumulation is an active process that allows the thyroid to concentrate iodide to at least 25 times its concentration in plasma. The iodide is converted to thyroglobulin that is stored as colloid droplets. These droplets then fuse with lysosomes, where the colloid is proteolyzed, releasing thyroxine and triiodothyronine. In PCB-treated rats, the uptake of iodide and synthesis of thyroglobulin appear to proceed normally, but fusion of the lysosomes and colloid droplets is inhibited (Collins et al., 1977; Collins and Capen, 1980a,b). This results in decreased secretion and lowered plasma levels of thyroid hormones, causing perturbations in the homeostatic mechanisms responsible for energy balance and producing many symptoms of PCB intoxication (see Thyroid Effects in Chapter V).

Metabolism

The role of metabolism in PCB toxicity has not been well defined and may vary with the endpoint observed. Enzyme induction is positively correlated with degree of chlorination and, thus, inversely correlated with rate of metabolism (see Metabolism in Chapter III). Toxicity in cell cultures is inversely correlated with the degree of chlorination and, thus, directly correlated with the rate of metabolism (Hoopingarner et al., 1972). It, therefore, appears that enzyme induction may be a function of the parent compound, while cellular toxicity is due to the production of more highly toxic metabolites.

Stadnicki and Allen (1979) studied the toxicity of 2,2',5,5'-tetrachlorobiphenyl and its metabolites, 2,2',5,5'-tetrachlorobiphenyl-4-ol and 2,2',5,5'-tetrachlorobiphenyl-3,4-oxide, in L-929 and HeLa cells. Both metabolites were more toxic than the parent PCB, with the oxide displaying the greatest degree of toxicity. This study, therefore, supports the involvement of metabolism in the cellular toxicity of PCBs.

Estrogenic activity has been observed in PCBs with low degrees of chlorination (see Reproductive Toxicity) and that are, therefore, rapidly metabolized. Since estrogenic activity appears to require the presence of two properly spaced hydroxy groups, the estrogenic activity of these PCBs is probably due to a metabolite rather than to the parent PCB.

The carcinogenic effects of PCBs (see Carcinogenicity in Chapter V) are probably due to a metabolite rather than the parent molecule. The critical event in the induction of tumors by chemical carcinogens is believed to be the formation of carcinogen-DNA adducts in the target cells. These adducts are formed when the PCBs are metabolized to an epoxide, which is an electrophilic species that can react with nucleophilic sites on cellular macromolecules (see Covalent Binding).

Interactive Effects

Many of the interactive effects between PCBs and other substances may be predicted from a knowledge of the enzyme inductive effects of PCBs. PCB pretreatment resulted in increased metabolism and excretion of pentobarbital and decreased pentobarbital induced sleeping times (Chu et al., 1977; Villeneuve et al., 1972), increased mutagenicity of benzo[a]pyrene (Hutton et al., 1979) and increased halothane and vinylidene fluoride hepatotoxicity (Sipes et al., 1978; Conolly et al., 1979). Thus, PCB pretreatment may result in either increased or decreased toxicity of other substances, depending on the role of oxidative metabolism in the induction of the toxic effect.

Cadmium has been demonstrated to have an antagonistic effect on the biochemical changes induced by PCBs (Suzuki, 1980; Horio et al., 1981). The exact nature of this interaction has not been elucidated; however, cadmium appears to inhibit the enzyme systems induced by PCBs.

Increasing dietary ascorbic acid levels have been demonstrated to reduce some, but not all, of the toxic effects of PCBs in rats and guinea pigs. Chakrabortz et al. (1978) reported that dietary ascorbic acid supplementation protected against alterations in enzyme activity and hepatic histopathology, but not the growth reduction observed in PCB treated rats. Kato et al. (1981) reported suppression of PCB-induced increases in serum cholesterol and phospholipid levels and hepatic lipid peroxidation, but not serum cortisol and triglyceride levels, in guinea pigs fed diets supplemented with 2000 mg ascorbic acid/kg diet. The mechanism of this interaction is not known, but Kato et al. (1981) proposed that ascorbic acid, functioning as an antioxidant, inhibits lipid peroxidation, thus reducing many of the toxic effects of PCBs.

PCBs also have mixed effects on tumor development. Anderson et al. (1983) administered 500 mg Aroclor 1254/kg bw to pregnant Swiss CD-1 mice on gestation day 19, then administered 5 mg N-nitrosodimethylamine/kg bw to the offspring on postnatal days 4 or 15. PCB treatment exerted a significant protective effect on the development of lung tumors in both sexes at 18 months and liver tumors in males at 28 weeks, but significantly increased the percentage of mice (both sexes) with liver tumors at 18 months.

PCBs have also been demonstrated to promote the development of enzyme-altered foci or hyperplastic nodules following administration of nitrosamines (Oesterle and Deml, 1983; Pereira et al., 1982) or N-2-fluorenylacetamide (Tatematsu et al., 1979). Since the PCB was administered after the initiating agent in these studies, this effect is likely due to a direct promotional action of the PCB rather than to alterations of metabolism.

Since PCBs are inhibitors of the immune system, PCB treatment would be expected to enhance the growth of transplantable tumors; however, only the opposite effect has been observed in studies to date (Kerkvliet and Kimeldorf, 1977a,b). Aroclor 1254 produced a dose-related inhibition of the growth of Walker 256 carcinosarcoma in Sprague-Dawley rats. The same inhibition was observed whether the PCB was administered 5 days before or after inoculation with tumor cells. The mechanism of this inhibition has not been elucidated.

Summary

Nine mechanisms (enzyme induction, inhibition of mitochondrial respiration, estrogenic effects and altered steroid metabolism, mobilization and redistribution of stored PCBs, covalent binding to cellular macromolecules, altered calcium metabolism, immune suppression, decreased thyroglobulin proteolysis, and metabolism) have been implicated in the induction of toxic effects by PCBs. Enzyme induction has been demonstrated in a wide variety of species. The enzymes induced by PCBs may be typified as either 3-methylcholanthrene type (P-448) or phenobarbital type (P-450). Minimum structural requirements for P-450 induction are symmetrical substitution at the ortho and para positions. P-448 induction requires substitution at the para and meta, but not ortho, positions (Goldstein et al., 1977). The induction of P-448 appears to correlate with the long-term toxicity of PCBs within a particular species (Yoshimura et al., 1979), but this correlation does not extend to interspecies comparisons (Shull et al., 1982).

PCBs may either inhibit or uncouple mitochondrial respiration. There is some evidence that the dominant effect depends on both the degree of chlorination and the concentration of PCB (Sivalingan et al., 1973; Khan and Cutkomp, 1982), but the mechanisms involved have not been elucidated.

PCBs have a number of effects on reproduction. These effects may result from a direct estrogenic effect of PCBs or from altered metabolism of steroid hormones secondary to enzyme induction. Bitman and Cecil (1970) have found that PCBs with <54% chlorination have weak estrogenic activity. PCB treatment has also been demonstrated to induce the metabolism and lower the metabolism of progesterone, estradiol-17_{φ}, and testosterone (Orberg and Ingvast, 1977; Derr and Dekker, 1979; Conney et al., 1967; Kintzman et al., 1977).

PCBs are highly lipophilic compounds that may be stored in body fat for long periods of time. During periods when body fat depots are mobilized, such as starvation, these stored PCBs may be redistributed, resulting in a delayed toxic effect.

Metabolism of PCBs may produce an electrophilic species (epoxide) that can bind covalently to cellular macromolecules. PCBs have been observed to bind to protein, RNA and DNA in microsomal metabolizing systems (Wyndham and Safe, 1978; Shimada and Sato, 1980). Binding to cellular DNA may, thus, be the mechanism by which PCBs induce their mutagenic and carcinogenic effects.

Kirkachi and Masuda (1973) observed accumulations of calcium in the heart and kidney tissue of individuals chronically poisoned with PCBs. One study in rats (Yagi et al., 1976) has also indicated the potential of PCBs to alter calcium metabolism *in vivo*.

Suppression of the immune system is one of the more sensitive indicators of PCB intoxication. Conradt et al. (1979) have demonstrated an inhibition of RNA synthesis in macrophages exposed to Aroclor 1242. They suggested that this inhibition may account for the immunosuppressive action of PCBs, since RNA synthesis is suspected to be involved in the acquisition and transfer of immunity.

PCB treatment produces numerous alterations in the structure and function of the thyroid gland, thus altering the homeostatic mechanisms regulating energy balance in vivo. The exact mechanism responsible for these effects is not known, but the effect appears to occur at the point where lysosomes fuse with colloid droplets (Collins et al., 1977, 1980a; Collins and Casper, 1980). When this fusion is blocked, thyroglobulin cannot be proteolyzed to thyroxine and tri-iodothyronine and collodial thyroglobulin accumulates in the follicular cells.

PCBs produce a wide variety of toxic responses, some of which appear to be due to the parent compound and some of which are due to metabolites. Enzyme induction is probably a function of the parent compound, since induction is greater following exposure to more highly chlorinated, thus more slowly metabolized, PCBs. Cellular toxicity, estrogenic activity and carcinogenicity are more likely to be the result of the formation of toxic metabolites.

PCBs also alter the effects of many other compounds. Many of these effects are the result of altered metabolism caused by the enzyme inductive effects of PCBs (Chu et al., 1977; Villeneuve et al., 1972; Hutton et al., 1979; Sipes et al., 1978; Conolly et al., 1979). Other interactions are not as well understood. Cadmium exerts an antagonistic effect on many aspects of PCB toxicity, apparently by inhibiting the enzyme systems induced by PCBs (Suzuki, 1980; Horio et al., 1981). Dietary ascorbic acid supplementation reduces some of the toxic effects of PCBs, possibly by inhibiting lipid peroxidation through its role as an antioxidant (Kato et al., 1981; Chakrabortz et al., 1978). PCBs also inhibit the growth of transplantable tumors (Anderson et al., 1983; Oesterle and Deml, 1983; Pereira et al., 1982), but the mechanism of this interaction is unknown.

VIII. QUANTIFICATION OF TOXICOLOGIC EFFECTS

The quantification of toxicological effects of a chemical consists of an assessment of the noncarcinogenic and carcinogenic effects. In the quantification of noncarcinogenic effects, an acceptable daily intake (ADI) is calculated. An adjusted acceptable daily intake (AADI) and health advisory (HA) values for the chemical are then calculated to define the appropriate drinking water concentrations to limit human exposure. For ingestion data, this approach is illustrated as follows:

$$\text{ADI} = \frac{\text{(NOAEL or LOAEL in mg/kg/day) (body weight in kg)}}{\text{Uncertainty/Safety Factor}} = \text{mg/day}$$

$$\text{AADI} = \frac{\text{ADI}}{\text{Drinking water volume in } \ell\text{/day}} = \text{mg/}\ell$$

where:

NOAEL = no-observed-adverse-effect level
LOAEL = lowest-observed-adverse-effect level
Body weight = 70 kg for adult or 10 kg for child
Drinking water volume = 2 ℓ/day for adults or 1 ℓ/day for children
Uncertainty/Safety Factor = 10, 100 or 1000

Utilizing these equations, the following drinking water concentrations are developed for noncarcinogenic effects:

1. A 1-day HA for 10 kg child
2. A 1-day HA for 70 kg adult
3. A 10-day HA for 10 kg child
4. A 10-day HA for 70 kg adult
5. A lifetime AADI for a 70 kg adult

The distinctions made between the HA calculations (items 1-4) are associated with the duration of anticipated exposure. Items 1 and 2 assume a single acute exposure to the chemical. Items 3 and 4 assume a limited period of exposure (possibly 1-2 weeks). The HA values will not be used in establishing a drinking water standard for the chemical. Rather, they will be used as informal scientific guidance to municipalities and other organizations when emergency spills or contamination situations occur. The AADI value (item 5) is intended to provide the scientific basis for establishing a drinking water standard based upon noncarcinogenic effects.

A NOAEL or LOAEL is determined from animal toxicity data or human effects data. For animal data, this level is divided by an uncertainty factor because there is no universally acceptable quantitative method to extrapolate from animals to humans. The possibility must be considered that humans are more sensitive to the toxic effects of chemicals than are animals. For human data, an uncertainty factor is also used to account for the heterogeneity of the human population in which persons exhibit differing sensitivities to toxic chemicals. An Office of Drinking Water (ODW) modification of the guidelines set forth by the National Academy of Sciences (NAS, 1977, 1980) are typically used in establishing uncertainty factors as follows:

- An uncertainty factor of 10 is used when good acute or chronic human exposure data are available and supported by acute or chronic toxicity data in other species.
- An uncertainty factor of 100 is used when good acute or chronic toxicity data identifying NOEL/NOAEL are available for one or more species, but human data are not available.
- An uncertainty factor of 1000 is used when limited or incomplete acute or chronic toxicity data are available or when only the acute or chronic toxicity data identify a LOAEL (but not NOEL/NOAEL) for one or more species are available.

The uncertainty factor used for a specific risk assessment is judgmental. Factors that cannot be incorporated in the NAS/ODW guidelines (NAS, 1977) for selection of an uncertainty factor, but must be considered, include the following: 1) the quality of the toxicology data, 2) the significance of the adverse effect, and 3) the existence of counterbalancing beneficial effects.

If toxicological evidence requires the chemical to be classified as a potential carcinogen, mathematical models are used to calculate the estimated excess cancer risks associated with the ingestion of the chemical by drinking water. The bioassay data used in these estimates are from animal experiments. In order to predict the risk for humans, these data must be converted to an equivalent human dose. This conversion includes correction for noncontinuous animal feeding, nonlifetime studies and for the difference in size. The factor that compensates for the size difference is the cube root of the ratio of the animal and human body weights. It is assumed that the average human body weight is 70 kg and that the average human consumes 2 ℓ of water/day. The multistage model is then fit to the equivalent human data to estimate the risk at low doses. The upper 95% confidence limit of this estimate is used. Excess cancer risks can also be estimated using other models such as the one-hit model, the Weibull model, the logit model and the probit model. There is no basis in the current understanding of the biological mechanisms involved in cancer to choose among these models. The estimates of low doses for these models can differ by several orders of magnitude.

The scientific data base used to calculate and support the setting of risk rate levels has an inherent uncertainty. This is because the tools of scientific measurement, by their very nature, involve both systematic and random error. In most cases, only studies using experimental animals have been performed. There is thus uncertainty when the data are extrapolated to humans. When developing risk rate levels, several other areas of uncertainty exist, such as 1) incomplete knowledge concerning the health effects of contaminants in drinking water, 2) the impact of test animal age, sex and species and the nature of target organ systems examined on the toxicity study results and 3) the actual rate of exposure of internal targets in test animals or humans. Dose-response data are usually only available for high levels of exposure, not for the lower levels of exposure for which a standard is being set. When there is exposure to more than one contaminant, additional uncertainty results from a lack of information about possible synergistic or antagonistic effects.

Existing Guidelines, Recommendations and Standards

Manufacture, sales and distribution of PCBs have been restricted under Section 6(e) of the Toxic Substances Control Act (TSCA) (P.L. 94-469). PCBs were restricted to sealed systems as of 1977, and manufacture and distribution were banned in 1979. Rules for the disposal of PCBs were proposed in 1978 (43 FR 7150).

The U.S. EPA (1980a) has set ambient water quality criteria for PCBs for the protection of humans from increased risk of cancer over the lifetime of 10^{-5}, 10^{-6} and 10^{-7} at 0.79, 0.079 and 0.0079 ng/ℓ. As a result of the large BCF, this criteria applies regardless of whether exposure occurs through consumption of 2 ℓ of water and 6.5 g of fish/day or through consumption of fish alone. The Food and Drug Administration (FDA) has set temporary tolerances for PCBs in food and food related products as indicated in Table VIII-1.

Occupational exposure limits have been recommended by the American Conference of Governmental Industrial Hygienists (ACGIH, 1980), and criteria set by the National Institute for Occupational Safety and Health (NIOSH, 1977) for PCBs in the workroom air. The TWA and STEL for Aroclor 1254, respectively, is 0.5 and 1.0 mg/m³, and 1 and 2 mg/m³ for Aroclor 1242 (ACGIH, 1980). The NIOSH (1977) criteria is 1.0 µg/m³ for a 10 hours/day, 40 hours/week exposure.

The NAS (1980) has developed a 24-hour SNARL for PCBs of 350 µg/ℓ based on the induction of mixed-function oxidase enzymes in the liver of rats administered Aroclor at doses of 1-2 mg/kg. For this analysis, an uncertainty factor of 100 was used, since only enzyme induction was reported in this dose range.

Noncarcinogenic Effects

Tests of the acute lethality of PCB products in laboratory animals, with the exception of the guinea pig, suggest that, in general, PCB products have similar toxicity regardless of route of administration, species or age of animal. The single dose oral LD_{50} value in rats, rabbits, mice and mink range from 0.5-20 g/kg bw (Grant and Phillips, 1974; Bruckner et al., 1973; Kimbrough et al., 1978; Fishbein 1974; Garthoff et al., 1981, Aulerich and Ringer, 1977). Route of administration also had little effect (<1 order of

TABLE VIII-1

FDA Regulations for PCBs*

Commodity	Temporary Tolerances (ppm)
Milk (fat basis)	1.5
Manufactured dairy products (fat basis)	1.5
Poultry (fat basis)	3.0
Eggs	0.3
Finished animal feeds	0.2
Animal feed components of animal origin	2.0
Edible portion of fish and shellfish	5.0
Infant and junior foods	0.2
Paper food packaging material	10.0

*Source: 21 CFR. 109.30. 51,725

magnitude) on lethality with the lethal dose for dermal administration in rabbits ranging from 0.7-3 g/kg bw (Nelson et al., 1972); while the lethal dose in mice administered PCBs by interperitoneal injection, ranged from 0.8-1.2 g/kg bw (Lewin et al., 1982).

There are only two indications of major differences in the acute toxicity of PCBs. First, there is limited evidence that the guinea pig may be more sensitive to the lethality of PCBs than other species. Miller (1944) observed a 100% mortality in a small group of guinea pigs receiving two oral doses of PCB (43% chlorine) at levels of 67 mg/animal at an interval of 7 days apart; and McConnell and Kinney (1978) reported a LD_{50-30} of 0.5 mg/kg for the PCB isomer 3,4,5-sym-HCB in guinea pigs. This indication of possible large inter-species differences in sensitivity is of concern in species-to-species extrapolation when there is insufficient data to indicate which experimental animal most accurately reflects the sensitivity of humans. The second problem concerns the possible large difference in toxicity of specific isomers of PCBs. There is limited data available from Biocca et al. (1981) that indicates that four different hexa-PCBs differ in LD_{50} value from 19 mg/kg to >64 mg/kg after oral administration to mice. It would not be unreasonable to assume that even larger differences will be encountered as more isomers are tested. Variation in toxicity of the different isomers is not of great concern in defining acceptable environmental levels, since individual isomers were not commercially made and released to the environment. The analytical methods used to measure environmental PCBs determines the level in mg of specific Aroclors. Since the Aroclors do not differ greatly in acute toxicity as described above, using data from the most toxic Aroclor should be protective without overly penalizing the other Aroclors. For the purpose of risk assessment, only studies using PCB products will be discussed.

There is some data available on the nonlethal acute toxicity of PCBs administered by the oral route for periods of 30 days or less. The effects described in these studies were alterations of the liver, thyroid and reproductive system. Rosin and Martin (1983) reported that a dose of 500 mg/kg of Aroclor 1254 for 14 days to CD-1 mice decreased pentabarbital sleeping time, indicating a substantial induction of microsomal enzymes. At lower doses, Sanders et al. (1974) reported that exposure of ICR mice to diets containing 250 or 62.5 ppm of Aroclor 1254 for 14 days resulted, respectively, in hepatomegaly and elevated serum corticosterone (the latter presumably as a result of altered liver steroid metabolism). These exposures would result in doses of 32.8 and 8.1 mg/kg bw/day, assuming a mouse consumes 13% of its body weight per day.

Similarly, Narbonne (1979) reported decreases in phenobarbital sleeping time in Sprague-Dawley rats maintained for 8 days on a diet containing 100 ppm of Phenoclor DP6 (dose of 5 mg/kg bw/day, assuming a rat consumes 5% of its body weight/day). PCB-induced increases in liver enzymes was suggested as the reason for the increase in testicular acid phosphatase observed by Dikshith et al. (1975) in Sprague-Dawley rats fed Aroclor 1254 at a dose of 50 mg/kg bw/day for 7 days. Increases in liver-to-body weight ratio appear to be one of the sensitive indicators of PCB exposure. Carter and Mercer (1983) reported that 64 mg/kg bw/day of Aroclor 1254 caused increased liver weight; while Grant and Phillips (1974) observed increased liver weight at doses as low as 12.5 and 5 mg/kg bw/day in male and female Wistar rats, respectively, receiving Aroclor 1254 in the diet for 14 days. Carter (1983) observed hepatomegaly in rats ingesting diets containing as little as 20 ppm Aroclor 1254 (1 mg/kg bw/day) for 14 days. Doses as low as 1 ppm of diet for 28 days caused liver microabcesses and an increased liver weight in 18-20 g 5-week-old C57B1/6J mice (Biocca et al., 1981). A dietary concentration of 0.3 ppm resulted in an increased liver weight with no other adverse effects (Biocca et al., 1981). This dose could be considered a NOAEL.

Besides changes in the liver, other effects reported for exposure to low levels of PCBs were increased thyroid activity in Sherman rats maintained on diets containing 250 ppm of Aroclor 1254 (12.5 mg/kg bw) for 14 days; in Sprague-Dawley rats, administration of Aroclor 1254 by gavage for 21 days at a dose of 0.05 g/kg bw/day resulted in weight loss and decreased body temperature (Komives, 1979; Komives and Alayoku, 1980). Enlarged thyroid has also been found in Osborne-Mendel rats maintained on diets containing 5 ppm of Aroclor 1254 (0.25 mg/kg bw/day) for 4 weeks (Collins and Capen, 1980b). This exposure level also resulted in increased liver enzymes in Holtzman rats (Garthoff et al., 1977).

The toxicity of PCBs resulting from exposures of between 30 and 90 days has been more extensively studied. Alterations in liver histopathology occurred as doses as low as 5 ppm diet for 5 weeks in Holtzman rats (Kasza et al., 1978b). In the mouse (MNRI) a dose of Clophen A-60, as low as 0.025 mg/mouse (0.8 mg/kg bw/day, assuming a mouse weight 0.03 kg) for 62 days, increased the estrous cycle, probably as a result of PCB-induced changes in liver steroid metabolism (Orberg and Kihlstrom, 1973). At higher dietary concentrations of 167 ppm (22 mg/kg bw) for 6 weeks, Aroclor 1016 and 1242 decreased the immunologic capability of BALB/CJ mice (Loose et al., 1978a).

Although other species have been tested to a lesser extent for this duration, the LOEL in these species were similar to those described for rats and mice. Rabbits exposed to diets containing 3 ppm of Aroclor 1254 (0.15 mg/kg bw/day, assuming a rabbit consumes 4.9% of its body weight per day) for 8 weeks developed hepatomegaly and immunosuppression. In the guinea pig, Vos and van Genderen (1973) reported that diets containing 250 ppm of Clophen A-60 (7 mg/kg bw/day, assuming a guinea pig consumes 2.8% of its body weight per day) for 4-7 weeks was lethal; while diets containing 50 ppm Clophen A-60 or Aroclor 1260 (1.4 mg/kg bw) for 4-7 weeks produced immunosuppression. Allen et al. (1974a) and Allen (1975) observed comedones and facial edema in rhesus monkeys ingesting diets containing 25 ppm Aroclor (1.1 mg/kg bw, assuming a monkey consumes 4.2% of its body weight per day) for 2 months. The LOELs observed in these species were slightly higher than the LOELs reported in rats and mice.

Studies of subchronic and chronic exposure (i.e., $\geq$90 days) to PCBs have failed to use sufficiently low doses to define a NOAEL in rats. In Sprague-Dawley rats, Allen et al. (1976) and Allen and Abrahamsen (1979) reported that a 52-week exposure to diets containing 100 ppm of Aroclor 1248, 1254 or 1262 (5 mg/kg bw/day) followed by a 13-week observation period resulted in hepatomegaly and liver necrosis. At a lower exposure of 75 ppm in the diet (3.75 mg/kg bw/day) for 36 weeks, Sprague-Dawley rats developed focal necrosis (Jonssen et al., 1981). Elevated liver porphyrine levels were detected by both Kimbrough et al. (1972) and Zinkl (1977) after exposure of Sherman rats for 8 months to 20 ppm Aroclor 1254 or 1260 (1 mg/kg bw/day) or CD rats for 20 weeks to 10 ppm of Aroclor 1254 (0.4 mg/kg bw/day). In a study employing near lifetime exposure (2 years), Morgan et al. (1981) reported an and Allen, 1975; Allen and Barsotti, 1976; Allen et al., 1980; Becker et al., 1979). These effects were observed at the lowest doses tested in the above studies that ranged from 2.5-3 ppm in the diet (0.095-0.126 mg/kg bw/day). In addition, Becker et al. (1979) reported that monkeys fed diets containing 3 ppm of PCBs had gastric lesions, body weight loss and reduced hemoglobin and leukocytes. In the monkey, doses as low as 0.1 mg/kg bw/day produced frank toxic effects; no studies have been conducted from which a NOAEL can be derived or to indicate how close 0.1 mg/kg bw/day is to the NOAEL for monkeys.

Although PCBs have not been demonstrated to be animal teratogens following oral exposure, these compounds have been demonstrated to adversely affect reproduction. When administered to pregnant Wistar rats at a dose of 100 mg/kg bw/day on days 6-15 of gestation, Villeneuve et al. (1971) observed no adverse effects; however, using the same treatment schedule, Spencer (1982) reported that Sprague-Dawley rats were infertile after receiving 15 mg/kg bw/day, that animals receiving 5 mg/kg bw/day had reduced litter weights and that 2.5 mg/kg bw/day was the NOEL. Rabbits had resorptions, abortions and dead fetuses at similar dose levels of 12.5 mg/kg bw/day administered on days 0-28 of gestation; however, slightly smaller doses of 10 mg/kg bw/day were reported to be the NOEL. The Hartly guinea pig, which has been shown to have greater sensitivity to the toxicity of PCBs than most other species, had macerated fetuses after receiving 2.2 mg/day (6.5 mg/kg bw/day) of Clophen A-50 on days 10-60 of gestation (Brunstroem et al., 1982).

Effective doses of PCBs were lower when exposure occurred before and during gestation. In a 2-generation study, Sherman rats maintained on diets containing 20 ppm Aroclor 1254 (1 mg/kg bw/day) had reduced litter size, and at 100 ppm (5 mg/kg bw/day) the pups that were born died during nursing (Linder et al., 1974). In this study, 5 ppm (0.25 mg/kg bw/day) was the NOEL. Complete loss of fertility was observed in male and female Wistar rats caged together for 9 weeks while ingesting 6.4 mg/kg bw/day of Aroclor 1254 emulsified in their drinking water (Baker et al., 1977). Males regained normal fertility after removal from treatment for 2 weeks. When Aroclor 1254 was administered to lactating Holtzman rats at 32 mg/kg bw/day on days 3, 5 and 7 of lactation, the future mating behaviour of nursing male pups was adversely affected (Sager, 1983). A lower dose of 8 mg/kg bw/day was a NOEL.

The mink and the monkey are the most sensitive species tested to the reproductive toxicity of the PCBs. Bleavins et al. (1980) maintained mink on diets containing 5 ppm Aroclor 1242 or 20 ppm Aroclor 1016 (doses of 0.75 and 3 mg/kg bw/day, assuming a mink consumes 15% of its body weight per day) for 18 months and observed complete reproductive failure in the Aroclor 1242 group and 25% mortality and infertility in the Aroclor 1016 group. In a limited study (8 animals/group), Allen et al. (1980) maintained rhesus monkeys on diets containing 2.5 or 5 ppm (0.1 or 0.2 mg/kg bw/day) of Aroclor 1248 for 18 months. In the low-dose group, increased abortions were observed; while in the high-dose group, the mothers showed overt signs of toxicity and no live births occurred. After removal from exposure for 1 year, fertility had still not returned to normal and some pups died during nursing. It is apparent that frank effects in reproduction were observed in monkeys at lower doses than the NOEL in rats, rabbits and guinea pigs following repeated exposure to PCBs.

Quantification of Noncarcinogenic Risk

Studies of PCB toxicity in experimental animals have demonstrated a progression of toxicologic responses correlated with dose for studies of 1-30 days. Carter (1983) observed hepatomegaly in rats ingesting PCBs at levels of 1 mg/kg bw/day (14 days). Carter (1983) exposed 10 groups of five animals each to diets containing 20 ppm of Aroclor 1254. At days 1, 2, 4, 8 and 14, animals were killed and body weight and liver weight determined. There was no statistically significant change in body weight; however, liver weights increased with duration of treatment as compared with controls. The increased liver weights were statistically significant (p=0.01) at days 4, 8 and 14 of exposure. Higher doses of ~5 mg/kg bw/day of Penochlor DP6 in rats and ~8mg/kg bw/day of Arochlor 1254 in mice resulted, respectively, in decreased barbitol sleeping time (8 days exposure) (Narbonne, 1979), and increased serum corticosterone (14-day exposure) mice (Sanders et al., 1974). This progression of effects suggests that PCB alterations of the liver may be affecting the animals ability to metabolize both endogenous compounds, steroids, as well as xenobiotics at dose levels of ~5 mg/kg bw/day. However, lower doses in slightly longer studies also show effects. For example, an estimated dose of 0.25 mg/kg bw/day for 4 weeks causes enlarged thyroid (Collins and Capen, 1980b), or an increase in liver enzymes (Garthoff et al., 1977). Dietary administration of 1.0 ppm for 5 weeks (~0.13 mg/kg bw/day assuming a mouse eats 13% of its body weight in food per day) caused an increase in liver weight associated with liver microabcesses in young C57Bl/6J mice (Biocca et al., 1981). A NOAEL of 0.3 ppm diet (0.039 mg/kg bw/day) was also apparent in this latter study.

Considering this data, a 1-day HA could be derived using the NOAEL of 0.039 mg/kg bw/day obtained for mice. This NOAEL is lower than the doses of PCBs that have been demonstrated to adversely affect reproduction in the rat (2.5 mg/kg bw/day) (Spencer, 1982), and the rabbit (12.5 mg/kg bw/day) (Villeneuve et al., 1971) when administration was for a substantial portion of gestation, and is lower than an estimated dose of 0.13 mg/kg bw/day in mice that caused an increase in liver weight associated with liver microabcesses. However, reproductive toxicity in the guinea pig and monkey, which appear to be very sensitive species in acute (lethal) and chronic studies, respectively, have not been adequately studied following short-term exposures. Moreover, PCBs are mixtures of isomers containing impurities, making it is very difficult to associate adverse health effects with specific forms of PCBs (Arochlors, Clophen, Kanechlor, etc.). Thus, a 1-day HA cannot be developed with confidence.

In studies of 30-90 days duration that are appropriate for use in the estimation of a 10-day HA, increased liver weight and decreased immunoactivity were observed in rabbits ingesting ~2.1 mg/kg bw/day (Street and Sharma, 1975). A dose of 0.92 mg/kg bw/day appeared to be a NOEL. An enlarged thyroid with other adverse sequellie were seen at doses of 5 ppm in diet of rats for 5 weeks (~0.25 mg/kg bw/day) (Kasza et al., 1978a). However, because of the reasons stated under the 1-day HA, there exists insufficient data to recommend a 10-day HA.

In deriving a lifetime AADI, it is inappropriate to use a dose level that has been shown to produce frank effects in any species tested. The only NOEL available was from the study of Koller (1977) in which mice ingested PCBs doses of 0.46, 4.57 or 45.7 mg/kg bw/day. Pathologic changes were observed in the liver at the middle dose, and the low dose was considered a NOEL. In the monkey, however, Barsotti and Allen (1975), Allen and Barsotti (1976), Allen et al. (1980) and Becker et al. (1979) reported that PCB exposures as low as ~0.1-0.126 mg/kg bw/day for as long as 18 months produce edema, gastric lesions and loss of body weight, as well as interfering with reproductive ability.

However, there exists insufficient data for recommending an ADI for PCBs. PCBs are very poorly metabolized and accumulate in the body. A possibility exists that large enough dose maybe deposited in the body. This dose may later be bioavailable to cause reproductive disturbances.

Carcinogenic Effect

A number of short-term assays predictive of carcinogenic potential have been performed using the Aroclors and individual isomers of PCBs. Negative results have been obtained in the reverse mutation assay using *S. typhimurium* (Schoeny et al., 1979; Schoeny, 1982; Heddle and Bruce, 1977), and in the dominant lethal assay in rats (Green et al., 1975b). PCB products also did not produce chromosomal changes in *D. melanogaster* (Nilsson and Ramel, 1974) or in the sperm and bone marrow cells of rats (Green et al., 1975b; Garthoff et al., 1977; Dikshith et al., 1975). Wyndham et al. (1976), however, observed increases in reversion frequency in *S. typhimurium* exposed to 4-chlorobiphenyl and to a lesser extent with Aroclor 1221, while the more highly chlorinated 2,2',5,5'-tetrachlorobiphenyl and Aroclor 1254 were negative. The positive response was observed in one strain, TA1538, in the presence of a metabolic activation system derived from rabbit liver. In addition, a weak positive response was observed by Peakall et al. (1972) in an assay of chromosomal abberration in the embryos of ring doves fed Aroclor 1254. The variable data observed with PCBs is consistent with the poor response and lack of correlation with animal carcinogenicity data reported for many highly chlorinated compounds in short-term assay.

Early bioassays of PCBs were inadequate as a result of small group size or periods of exposure extending for <1 year. These studies failed to demonstrate that PCBs were carcinogenic when fed to rats or mice (Kimura and Baba 1973; Ito et al., 1973; Ito et al., 1974; Kimbrough and Linder, 1974). The study by Kimbrough and Linder (1974) in which 50 BALB/CJ male mice were fed diets containing 300 ppm Aroclor 1254 for 6 or 11 months was suggestive that Aroclor 1254 was a liver carcinogen. Of the 22 animals surviving PCB treatment for 11 months, all had areas of adenofibrosis in the liver, and seven had histologically identified hepatomas. In animals surviving 11 months which were maintained on PCB-contaminated diets for 6 months and in control animals, there were, respectively, only 0/24 and 0/58 livers with areas of adenofibrosis and 1/24 and 0/58 animals with hepatomas. There were no histologically identified hepatocellular carcinomas in any group.

In a later study in rats using a larger group size and a longer exposure period, Kimbrough et al. (1975) reported an increased incidence in hepatocellular carcinomas in animals exposed to PCBs. In this study, 200 female Sherman rats were exposed to diets containing a nominal 100 ppm (range 70-107 ppm) of Aroclor 1260 for 630 days. The incidence of hepatocellular carcinomas in the treated group compared with control animals was 26/184 and 1/173, respectively; while the incidence of hepatic neoplastic nodules were 144/184 and 0/173, respectively.

In the only other chronic bioassay performed (NCI, 1978), 24 male and 24 female Fischer 344 rats/group were maintained on diets containing 0, 25, 50 or 100 ppm Aroclor 1254. Although dose-related increases in nodular hyperplasia were observed, there was no significant increase in neoplastic lesions. With respect to the tumor incidence reported by Kimbrough et al. (1975), the number of rats/group in the NCI study may have been too small to detect a carcinogenic response.

There is very limited data on the carcinogenicity of PCBs in humans. In a survey of 1200 Yusho patients, 5.5 years after exposure to PCBs, 41% of the 22 deaths were attributed to neoplasia (Kuratsume, 1976; Urabe, 1974). The relevance of these findings is unclear since the tumors were at various and common sites, and comparable incidences for an unexposed population were not presented. In two letters to the editor, Bahn et al. (1976, 1977) described suspected increases in malignant melanomas in a small group of workers exposed to Aroclor 1254. In the 31 "heavily exposed" workers, there were two cases of melanoma; while in the 41 "less heavily" exposed workers, there was one melanoma. IARC (1978) estimates that only 0.04 cases would be expected in this number of individuals. The only epidemiology study (Davidorf and Knupp, 1979) examined the association between ocular melanomas and populations residing in areas of known high environmental levels of PCBs. The authors concluded that a causal relationship between PCBs and ocular melanomas was not demonstrated. IARC (1978) considered the association between exposure to Aroclor 1254 and malignant melanomas described in two letters to the editor of the New England Journal of Medicine (Bahn et al., 1976, 1977) to be suggestive evidence that PCBs are human carcinogens.

Quantification of Carcinogenic Risk

The U.S. EPA Cancer Assessment Group (CAG) has used the data from female rats in the study by Kimbrough et al. (1975) to quantify the carcinogenic risk from exposure to PCBs. In this analysis, the TWA concentration of PCB (Arochlor 1260) in the diet was determined to be 88.4 ppm that was associated with a daily dose of 4.42 mg/kg bw/day by assuming that an adult rat consumes food equal to 5% of its body weight per day. In addition, for this analysis, the incidence of hepatocellular carcinomas (26/184 in treated and 1/173 in controls) and neoplastic nodules (144/184 in treated and 0/173 in controls) were combined to produce tumor incidences of 170/184 and 1/173 in the treated and control groups, respectively. Using this data and the linearized multistage model a q_1* for human exposure to Arochlor 1260 of 4.3396 (mg/kg/day)$^{-1}$ was calculated using the data in Table VIII-2.

Using this q_1*, the concentration of Arochlor 1260 in water that would result in an increased lifetime risk level of 10^{-5} for a 70 kg man, assuming consumption of 2 ℓ of water/day, can be calculated as follows:

$$\text{Concentration in water} = 10^{-5} \times 70 \text{ kg} \div q_1^* \text{ (mg/kg/day)}^{-1} \div 2 \text{ ℓ}.$$

Thus, using the q_1* for humans of 4.3396 (mg/kg/day)$^{-1}$, the concentration of PCBs in water corresponding to lifetime risks of 10^{-4}, 10^{-5} and 10^{-6} are 0.8, 0.08 and 0.008 µg/ℓ, respectively. The adoption of these potency and concentration levels for all PCBs requires an assumption that all PCB compounds are carcinogenic and that the potency of Arochlor 1260 is representative of any mixture or any other PCB compound.

TABLE VIII-2

Data Used as the Basis for the q_1*[†]

Species	rat
Strain	Sherman
Sex	female
Body weight (assumed)	0.4 kg
Length of exposure	645 days
Length of experiment	730 days
Tumor site	liver
Tumor type	combined hepatocellular carcinomas and neoplastic nodules
PCB product tested	Aroclor 1260

Dose (mg/kg/day)	Incidence (No. responding/No tested)
0	1/173
4.42	170/184

[†] Source: Data from the study of Kimbrough et al., 1975

Special Consideration

High Risk Subpopulation. Two separate groups of high risk subpopulations for exposure to PCBs may be identified. The first group includes those persons with the potential for frequent or high exposure, namely, occupationally-exposed workers and breast-fed infants, as PCBs are excreted in the breast milk of lactating humans (Miller, 1977; Rogan et al., 1980; Wickizer et al., 1981; Mes and Davies, 1979; Kuwabara et al., 1979; Hofvander et al., 1981). The second group includes those persons with an inability to oxidize PCBs via glucuronidation to facilitate detoxification and elimination of these toxicants, such as embroyos, fetuses and neonates (2-3 months old) (Calabrese and Sorenson, 1977; Gillette, 1967; Nyhan, 1961), especially breast-fed infants who receive a steroid via human breast milk that inhibits glucuronyl transferase activity (Calabrese and Sorenson, 1977; Gartner and Arias, 1966), children simultaneously exposed to the antibiotic novobiocin (Lokietz et al., 1963; Calabrese and Sorenson, 1977), persons with Gilbert's syndrome or Crigler and Najjar syndrome (Lester and Schmid, 1964; Calabrese and Sorenson, 1977) or persons with hepatic infections such as infectious hepatitis (Calabrese and Sorenson, 1977).

Co-Carcinogenesis, Initiation and Promotion. DiGiovanni et al. (1978) demonstrated that Aroclor 1254 had weak tumor initiating activity in the mouse 2-stage tumorigenesis models. Promoting activity was not indicated for this Aroclor in a study by Berry et al. (1978). Using other experimental systems, Ito et al. (1973) observed an increased incidence of liver tumors in rats co-administered PCBs and benzene hexachloride as compared with benzene hexachloride treatment alone. Co-administration of other potent liver carcinogens, 3'-methyl-4-dimethylaminoazobenzene, N-2-fluorenylacetemide and diethylnitrosamine, with PCBs, however, has resulted in the inhibition of the tumorigenic response (Ito et al., 1973). Similarly, antineoplastic effects were reported by Nishizumi (1980) for pups of dams administered Kanechlor-500 and diethylnitrosamine. These studies make it apparent that exposure to PCBs can affect the carcinogenicity of other xenobiotics.

Summary

The recommended health advisories developed in this document are summarized in Table VIII-3. Health Advisories for 1-day and 10-day, and the AADI are not recommended at this time due to a deficient toxicity data base. A cancer based criteria was derived and calculated for excess lifetime cancer risks of 10^{-4}, 10^{-5} and 10^{-6}. The respective water concentrations are 0.8, 0.08 and 0.008 µg/ℓ.

TABLE VIII-3

Summary of Health Advisories and Lifetime Cancer Risk for PCBs*

Criteria	μg/ℓ	μg/kg bw/day
1-day HA (adult)	not recommended	--
1-day HA (child)	not recommended	--
10-day HA (adult)	not recommended	--
10-day HA (child)	not recommended	--
Lifetime AADI	not recommended	--
10^{-4} excess cancer risk	0.8	0.01
10^{-5} excess cancer risk	0.08	0.001
10^{-6} excess cancer risk	0.008	0.0001

*See text for derivation of HAs, AADI and lifetime cancer risk

IX. REFERENCES

ACGIH (American Conference of Governmental Industrial Hygienists). 1980. Documentation of the Threshold Limit Values, 4th ed. (Includes Supplemental Documentation, 1981). Cincinnati, OH. p. 486.

Albro, P.W. and L. Fishbein. 1972. Intestinal absorption of polychlorinated biphenyls in rats. Bull. Environ. Contam. Toxicol. 8: 26.

Allen, J.R. 1975. Response of the non-human primate to polychlorinated biphenyls exposure. Fed. Proc. 34: 1675.

Allen, J.R. and L.J. Abrahamson. 1973. Morphological and biochemical changes in the liver of rats fed polychlorinated biphenyls. Arch. Environ. Contam. Toxicol. 1: 265.

Allen, J.R. and L.J. Abrahamson. 1979. Responses of rats exposed to polychlorinated biphenyls for fifty-two weeks. II. Compositional and enzymic changes in the liver. Arch. Environ. Contam. Toxicol. 8: 191.

Allen, J.R. and D.A. Barsotti. 1976. The effects of transplacental and mammary movement of the PCBs on infant rhesus monkeys. Toxicology. 6: 331.

Allen, J.R. and D.H. Norback. 1973. Polychlorinated biphenyls- and triphenyl-induced gastric mucosal hyperplasia in primates. Science. 179: 498.

Allen, J.R., D.H. Norback and I.C. Hsu. 1974a. Tissue modifications in monkeys as related to absorption, distribution and excretion of polychlorinated biphenyls. Arch. Environ. Contam. 2: 86.

Allen, J.R., L.A. Carstens and D.A. Barsotti. 1974b. Residual effects of short-term, low-level exposure of non-human primates to polychlorinated biphenyls. Toxicol. Appl. Pharmacol. 30: 440.

Allen, J.R., L.A. Cartens, L.J. Abrahamson and R.J. Marlar. 1975. Responses of rats and non-human primates to 2,5,2'.5'-tetrachlorobiphenyl. Environ. Res. 9: 265.

Allen, J.R., L.A. Carsten and L.J. Abrahamson. 1976. Responses of rats exposed to polychlorinated biphenyls for 52 weeks. I. Comparison of tissue levels of PCB and biological changes. Arch. Environ. Contam. Toxicol. 4: 409.

Allen, J.R., W.A. Hargraves, M.T. Hsia, et al. 1979a. Comparative toxicology of chlorinated compounds on mammalian species. Pharmacol. Ther. 7(3): 513-147.

Allen, J.R., D.A. Barsotti, L.K. Lambrecht and J.P. Van Miller. 1979b. Reproductive effects of halogenated aromatic hydrocarbons on nonhuman primates. Ann NY Acad. Sci. 320: 419.

Allen, J.R., D.A. Barsotti and L.A. Carstens. 1980. Residual effects of polychlorinated biphenyls on adult nonhuman primates and their offspring. J. Toxicol. Environ. Health. 6(1): 55-66.

Alvares, A.P. and A. Kappas. 1979. Lead and polychlorinated biphenyls: Effects on heme and drug metabolism. Drug Metab. Rev. 10: 91-106.

Alvares, A.P., D.R. Bickers and A. Kappas. 1973. Polychlorinated biphenyls: A new type of inducer of cytochrome P-448 in the liver. Proc. Natl. Acad. Sci. 70: 1321.

Alvares, A.P., A. Fishbein, K.E. Anderson and A. Kappas. 1977. Alterations in drug metabolism in workers exposed to polychlorinated biphenyls. Clin. Pharmacol. Ther. 22: 140.

Anderson, L.M., K. Van Havere and J.M. Budinger. 1983. Effects of polychlorinated biphenyls on lung and liver tumors initiated in suckling mice by N-nitrosodimethylamine. J. Natl. Cancer Inst. 71(1): 157-163.

Anderson, M.W., T.E. Eling, R.J. Lutz, R.L. Dedrick and H.B. Matthews. 1977. The construction of a pharmacokinetic model for the disposition of polychlorinated biphenyls in the rat. Clin. Pharmacol. Ther. 22: 765.

Atlas, E., A. Velasco, K. Sullivan, C.S. Giam. 1983. A radiotracer study of air-water exchange of synthetic organic compounds. Chemosphere. 12: 1251-1258.

Aulerich, R.J. and R.K. Ringer. 1977. Current status of PCB toxicity to mink, and effect on their reproduction. Arch. Environ. Contam. Toxicol. 6: 279.

Aulerich, R.J., R.K. Ringer and S. Iwamoto. 1973. Reproductive failure and mortality in mink fed on Great Lakes fish. J. Reprod. Fert. (Suppl.) 19: 365.

Bahn, A.K., I. Rosenwaike, N. Herrmann, P. Grover, J. Stellman and K. O'Leary. 1976. Melanoma after exposure to PCBs. New Engl. J. Med. 295: 450.

Bahn, A.K., P. Grover, I. Rosenwaike, K. O'Leary and J. Stellman. 1977. PCB? and melanoma. N. Gig. J. Med. 296: 108.

Bailey, J., V. Knauf, W. Mueller and W. Hobson. 1980. Transfer of hexachlorobenzene and polychlorinated biphenyls to nursing infant rhesus monkeys: Enhanced toxicity. Environ. Res. 21(1): 190-196.

Baker, F.D., B. Bush, C.F. Tumasonis and F.C. Lo. 1977. Toxicity and persistence of low-level PCB in adult wistar rats, fetuses, and young. Arch. Environ. Contam. Toxicol. 5(2): 143-156.

Barsotti, D.A. 1981. Gross, clinical and reproductive effects of polychlorinated biphenyls (PCBs) in the rhesus monkey. Diss. Abstr. Int. 41(10): 3744-3745.

Barsotti, D.A. and J.R. Allen. 1975. Effects of polychlorinated biphenyls on reproduction in the primate. Fed. Proc. 34: 338.

Barsotti, D.A., R.J. Marlar and J.R. Allen. 1976. Reproductive dysfunction in rhesus monkeys exposed to low levels of polychlorinated biphenyls (Aroclor 1248). Food Cosmet. Toxicol. 14: 99-103.

Bastomsky, C.H. 1974. Effects of a polychlorinated biphenyls mixture (Aroclor 1254) and DDT on biliary thyroxin excretion in rats. Endocrinology. 95: 1150.

Baumann, M., E. Deml, E. Schaeffer and H. Greim. 1983. Effects of polychlorinated biphenyls at low dose levels in rats. Arch. Environ. Contam. Toxicol. 12(5): 509-515.

Becker, G.M., W.P. McNulty and M. Bell. 1979. Polychlorinated biphenyls-induced morphologic changes in the gastric mucosa of the rhesus monkey. Lab. Invest. 40: 373.

Bell, M. 1983. Untrastructural features of the murine cutaneous microvasculature after exposure to polychlorinated biphenyls compounds (PCBs) and benzo(a)pyrene (BAP). Virchows Arch. B. 42(2): 131-142.

Benthe, H.F., J. Knop and A. Schmoldt. 1972. Absorption and distribution of polychlorinated biphenyls (PCB) after inhalatory application. Arch. Toxicol. 29: 85.

Bercovici, B., M. Wassermann, S. Cucos, M. Ron, D. Wassermann and A. Pines. 1983. Serum levels of polychlorinated biphenyls and some organochlorine insecticides in women with recent and former missed abortions. Environ. Res. 30(1): 169-174.

Berczy, Z.S., L.M. Cobb and C.P. Cherry. 1974. Acute inhalation toxicity to the rat of decachlorodiphenyl. Huntingdon, England, Huntingdon Research Centre, March. (Cited in NIOSH, 1977)

Berlin, M., J. Gage and S. Holm. 1975. Distribution and metabolism of 2,4,5,2',4',5'-pentachlorobiphenyl. Arch. Environ. Health. 30(3): 141-147. CA 82: 150084n. (Cited in Callahan et al., 1979)

Berry, D.L., J.D. Giovanni, M.R. Juchau, W.M. Bracken, G.L. Gleason and T.J. Slaga. 1978. Lack of tumor-promoting ability of certain environmental chemicals in a two-stage mouse skin tumorigenesis assay. Res. Commun. Chem. Pathol. Pharmacol. 20: 101.

Bickers, D.R., L.C. Harber, A. Kappas and A.P. Alvares. 1972. Polychlorinated biphenyls. Comparative effects of high and low chlorine containing Aroclors on hepatic mixed function oxidase. Res. Commun. Chem. Pathol. Pharmacol. 3: 505.

Biocca, M., B.N. Gupta, K. Chae, J.D. McKinney and J.A. Moore. 1981. Toxicity of selected symetrical hexachlorobiphenyl isomers in the mouse. Toxicol. Appl. Pharmacol. 58: 461.

Bitman, J. and H.C. Cecil. 1970. Estrogenic activity of DDT analogs and polychlorinated biphenyls. J. Agric. Food Chem. 18: 1108.

Bleavins, M.R., R.J. Aulerich and R.K. Ringer. 1980. Polychlorinated biphenyls (Aroclors 1016 and 1242): Effects on survival and reproduction in mink and ferrets. Arch. Environ. Contam. Toxicol. 9(5): 627-635.

Block, W.D. and H.H. Cornish. 1959. Metabolism of biphenyl and 4-chlorobiphenyl in the rabbit. J. Biol. Chem. 234: 3301-3302.

Bowes, G.W., M.J. Mulvihill, B.R.T. Simoneit, A.L. Burlingame and R.W. Risebrough. 1975. Identification of chlorinated dibenzofurans in American polychlorinated biphenyls. Nature. 256: 305-307.

Bowman, R.E. and M.P. Heironimus. 1981. Hypoactivity in adolescent monkeys perinatally exposed to PCBs and hyperactive as juveniles. Neurobehav. Toxicol. Teratol. 3(1): 15-18.

Bowman, R.E., M.P. Heironimus and J.R. Allen. 1978. Correlation of PCB body burden with behavioral toxicology in monkeys. Pharmacol. Biochem. Behav. 9: 49.

Bowman, R.E., M.P. Heironimus and D.A. Barsotti. 1981. Locomotor hyperactivity in PCB-exposed rhesus monkeys. Neurotoxicology. 2(2): 251-268.

Brown, J.F., R.E. Wagner, D.L. Bedard, et al. 1984. PCB transformation in upper Hudson sediment. Northeastern Environ. Sci. 3(3-4): (In press)

Branson, D.R., G.E. Blau, H.C. Alexander and W.B. Neely. 1975. Bioconcentration of 2,2',4,4'-tetrachlorobiphenyl in rainbow trout as measured by an accelerated test. Trans. Am. Fish. Soc. 4:785-792. (Cited in Callahan et al., 1979)

Bruckner, J.V., K.L. Khanna and H.H. Cornish. 1973. Biological responses of the rat to polychlorinated biphenyls. Toxicol. Appl. Pharmacol. 24: 434.

Bruckner, J.V., K.L. Khanna and H.H. Cornish. 1974a. Effect of prolonged ingestion of polychlorinated biphenyls on the rat. Food Cosmet. Toxicol. 12: 323.

Bruckner, J.V., K.L. Khanna and H.H. Cornish. 1974b. Polychlorinated biphenyl-induced alteration of biologic parameters in the rat. Toxicol. Appl. Pharmacol. 28: 189.

Brunstroem, B., I. Kihlstroem and U. Lundkvist. 1982. Studies of fetal death and fetal weight in guinea pigs fed polychlorinated biphenyls (PCB). Acta. Pharmacol. Toxicol. 50(2): 100-103.

Burns, L.H., D.M. Cline and R.R. Lassiter. 1981. Exposure Analysis Modeling System (EXAMS). Prepared by Environmental Research Lab, Office of Research and Development, U.S. EPA, Athens, GA.

Calabrese, E.J. and A.J. Sorenson. 1977. The health effects of PCBs with particular emphasis on human high risk groups. Rev. Environ. Health. 2(4): 285-304.

Callahan, M.A., M.W. Slimak, N.W. Gabel, et al. 1979. Water-Related Environmental Fate of 129 Priority Pollutants, Vol. I, EPA 440/4-79-029a, Office of Water Planning and Standards, Office of Water and Wastewater Management, U.S. EPA, Washington, D.C.

Carlson, G.P. 1980. Influence of starvation on the induction of xenobiotic metabolism by polychlorinated biphenyls. Life Sci. 27(17): 1571-1576.

Carter, J.W. 1983. Onset of hepatomegaly in PCB (Aroclor 1254)-treated rats. Bull. Environ. Contam. Toxicol. 31(2): 183-187.

Carter, J.W. and I.L. Cameron. 1977. Sublethal effects of a pure polychlorobiphenyl on mice. Exp. Mol. Pathol. 26(2): 228-250.

Carter, J.W. and J. Clancy Jr. 1980. Acutely administered polychlorinated biphenyls (PCBs) decrease splenic cellularity but increase its ability to cause graft-versus-host reactions in BALB/c mice. Immunopharmacology. 2(4): 341-347.

Carter, J.W. and L.P. Mercer. 1983. Prediction of food intake, weight gain and organ weights in rats as a formulation of dietary arochlor 1254 (PCBs) by the saturation kinetics model for physiological responses. Nutr. Rep. Int. 27: 561-568.

Chakraborty, D., A. Bhattacharyya, J. Chatterjee, et al. 1978. Biochemical studies on polychlorinated biphenyls toxicity in rats: Manipulation by Vitamin C. Intern. J. Vit. Nutr. Res. 48: 22.

Chang, K.J., J.S. Cheng, P.C. Huang and T.C. Tung. 1980a. Study of patients with PCB poisoning. J. Formosan Med. Assoc. 79: 304-313. (Cited in Chang et al., 1981)

Chang, K-J., F-J. Lu, T-C. Tung and T-P. Lee. 1980b. Studies on patients with polychlorinated biphenyls poisoning. 2. Determination of urinary coproporphyrin, uroporphyrin, delta-aminolevulinic acid and porphobilinogen. Res. Commun. Chem. Pathol. Pharmacol. 30(3): 547-554.

Chang, K.-J., K.-H. Hsieh, T.-P. Lee, S.-Y. Tang and T.C. Tung. 1981. Immunologic evaluation of patients with polychlorinated biphenyl poisoning: Determination of lymphocyte subpopulations. Toxicol. Appl. Pharmacol. 61: 58-63.

Chen, P.H., J.M. Gaw, C.K. Wong and C.J. Chen. 1980. Levels and gas chromatography patterns of PCBs in blood of patients after PCB poisoning in Taiwan. Bull. Environ. Contam. Toxicol. 25: 325-329.

Chen, P.H., K.T. Chang and Y.D. Lu. 1981. Polychlorinated biphenyls and polychlorinated dibenzofurans in the toxic rice-bran oil that caused PCB poisoning in Taichung. Bull. Environ. Contam. Toxicol. 26(4): 489-495.

Chen, P.H., M.L. Luo, C.K. Wong and C.J. Chen. 1982. Comparative rates of elimination of some individual polychlorinated biphenyls from the blood of PCB-poisoned patients in Taiwan. Food Chem. Toxicol. 20(4): 417-425.

Chen, P.R. and H.B. Matthews. 1974. Metabolism and excretion of 2,4,5,2',5-pentachlorobiphenyl (PCB) in the male rat. Toxicol. Appl. Pharmacol. 29: 88.

Chen, P.R., J.D. McKinney and H.B. Matthews. 1976. Metabolism of 2,4,5,2',5'-pentachlorobiphenyl in the rat. Drug Metab. Disp. 4: 362-367.

Chiou, C.T., V.H. Freed, D.W. Schmedding and R.L. Kohnert. 1977. Partition coefficients and bioaccumulation of selected organic chemicals. Environ. Sci. Technol. 11(5): 475-678.

Chu, C.K., V.J. Stella, J.V. Bruckner and W.D. Jiang. 1977. Effects of long-term exposure to environmental levels of polychlorinated biphenyls on pharmacokinetics of pentobarbital in rats. J. Pharm. Sci. 66(2): 238-241.

Clegg, D.J. 1971. Embryotoxicity of chemical contaminants of food. Food Cosmet. Toxicol. 9: 195-205.

Collins, W.T. and C.C. Capen. 1980a. Biliary excretion of thyroxine-I-125 and fine structural alterations in the thyroid glands of gunn-rats fed PCBs. Lab Invest. 43: 158.

Collins, W.T. and C.C. Capen. 1980b. Ultrastructural and functional alterations of the rat thyroid gland produced by polychlorinated biphenyls compared with iodide excess and deficiency, and thyrotropin and thyroxine administration. Virchows Arch. B. 33(3): 213-231.

Collins, W.T. and C.C. Capen. 1980c. Fine structural lesions and hormonal alterations in thyroid glands of perinatal rats exposed in utero and by the milk to polychlorinated biphenyls. Am. J. Pathol. 99(1): 125-141.

Collins, W.T., C.C. Capen, L. Kasza, C. Carter and R.E. Dailey. 1977. Effect of polychlorinated biphenyls (PCB) on the thyroid gland of rats. Am. J. Pathol. 89: 119.

Conney, A.H., R.M. Welch, R. Kuntzman and J.J. Burns. 1967. Effects of pesticides on drug and steroid metabolism. Clin. Pharm. Therap. 8: 2-10.

Conolly, R.B., S. Szabo and R.J. Jaeger. 1979. Vinylidene fluoride: Acute hepatotoxicity in rats pretreated with PCB or phenobarbital. Proc. Exp. Biol. Med. 162: 163.

Conradt, P., W.F. Muller, L. Loose, et al. 1979. Incorporation of [^{3}H]Uridine into RNA under the influence of dieldrin and polychlorinated biphenyls. Ecotoxicol. Environ. Safety. 3: 10.

Curley, A., V.W. Burse and M.E. Grim. 1973. Polychlorinated biphenyls: Evidence of transplacental passage in the Sherman rat. Food Cosmet. Toxicol. 11: 471.

Cutkomp, L.J., H.H. Yap, D. Desaiah and R.B. Koch. 1972. The sensitivity of fish ATPases to polychlorinated biphenyls. Environ. Health Perspect. 1: 165-168.

Davidorf, F.H. and J.A. Knupp. 1979. Epidemiology of ocular melanoma. Incidence and geographic relationship in Ohio (1967-1977). Ohio State Med. J. 75(9): 561-564.

Deml, E. and D. Oesterle. 1982. Sex-dependent promoting effect of polychlorinated biphenyls on enzyme-altering islands induced by diethylnitrosamine in rat liver. Carcinogenesis. 3: 1449-1453.

Derr, S.K. 1978. In vivo metabolism of exogenous progesterone by PCB treated female rats. Bull. Environ. Contam. Toxicol. 19: 729.

Derr, S.K. and J. Dekker. 1979. Alterations of androgenicity in rats exposed to PCBs (Aroclor 1254). Bull. Environ. Contam. Toxicol. 21: 43.

Desaiah, D., L.K. Cutkomp, H.H. Yap and R.B. Koch. 1972. Inhibition of oligomycin-sensitive and insensitive magnesium adenosine triphosphatase activity in fish by polychlorinated biphenyls. Biochem. Pharmacol. 21: 857-865.

Dexter, R.N. and S.P. Pavlov. 1978. Mass solubility and activity coefficients of stable organic chemicals in the marine environment: Polychlorinated biphenyls. Mar. Chem. 6: 41-53.

DiGiovanni, J., T.J. Slaga, D.L. Berry and M.R. Juchan. 1978. The effects of 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD) and Aroclor 1254 on 7,12-dimethylbenz[A]-anthracene metabolism and tumor initiating activity in mouse skin. Proc. AACR, ASCO. 19: 110.

Dikshith, T.S.S., W. Rockwood, R. Abraham and F. Coulston. 1975. Effects of polychlorinated biphenyls (Aroclor 1254) on rat testis. Exp. Mol. Pathol. 22: 376.

Doskey, P.V. and A.W. Andren. 1981. Modeling the flux of atmospheric polychlorinated biphenyls across the air/water interface. Environ. Sci. Technol. 15: 705-711.

Drinker, C.K., M.F. Warren and G.A. Bennett. 1937. The problem of possible systemic effects from certain chlorinated hydrocarbons. J. Ind. Hyg. Toxicol. 19: 283-311.

Dunn, J.P., J.W. Carter and D.A. Henderson. 1983. Effect of polychlorinated biphenyls (arochlor 1254) on rhythmic pituitary-adrenal function. Bull. Environ. Contam. Toxicol. 31: 322-325.

Ecobichon, D.J. 1975. The influence of polychlorinated biphenyl compounds on the hepatic function in the rat. Environ. Sci. Res. 7: 20.

Eisenreich, S.J., B.B. Looney and J.D. Thornton. 1981. Airborne organic contaminants in the Great Lakes ecosystem. Environ. Sci. Technol. 15: 30-38.

Felt, G.R., W.F. Mueller, M.J. Iatropoulos, F. Coulston and F. Korte. 1977. Chronic toxicity of 2,5,4'-trichlorobiphenyl in young rhesus monkeys. I. Body distribution, elimination and metabolism. Toxicol. Appl. Pharmacol. 41(3): 619-627.

Fischbein, A., M.S. Wolff, R. Lilis, J. Thornton and I.J. Selikoff. 1979. Clinical finding among PCB-exposed capacitor manufacturing workers. NY Acad. Sci. Ann. 320(May): 703.

Fishbein, L. 1974. Toxicity of chlorinated biphenyls. Ann. Rev. Pharmacol. 14: 139.

Fukuba, H. and A. Kanamori. 1982. Effects of non-nutritional polysaccharides on the excretion of polychlorinated biphenyls. Nutr. Rep. Int. 26(2): 281-287.

Fuller, G.B., V. Krauf, W. Mueller and W.C. Hobson. 1980. PCB augments LH-induced progesterone synthesis. Bull. Environ. Contam. Toxicol. 25: 65.

Funatsu, I., et al. 1971. A chlorobiphenyls induced fetopathy. Fukuoka Acta. Med. 62: 139. (Jap.) (Cited in Kuratsune et al., 1972)

Furukawa, K., K. Tonomura and A. Kamibayashi. 1978. Effect of chlorine substitution on the biodegradability of polychlorinated biphenyls. Appl. Environ. Microbiol. 35(2): 223-227. (Cited in Callahan et al., 1979)

Furukawa, K., N. Tomizuka, A. Kamikayash. 1983. Metabolic breakdown of kanechlors (polychlorobiphenyls and their products) of Acinetobacter sp. Appl. Environ. Microbiol. 46: 140-145.

Gage, J.C. and S. Holm 1976. The influence of molecular structure on the retention and excretion of polychlorinated biphenyls by the mouse. Toxicol. Appl. Pharmacol. 36: 555-560.

Gardner, A.M., J.T. Chen, J.A.G. Roach and E.P. Ragelis. 1973. Polychlorinated biphenyls: Hydroxylated urinary metabolites of 2,2',5,5'-tetrachlorobiphenyl identified in rabbits. Biochem. Biophys. Res. Commun. 55: 1377.

Garten, C.T., Jr. and J.R. Trabalka. 1983. Evaluation of models for predicting terrestrial food chain behavior of xenobioti. Environ. Sci. Technol. 17: 590-595.

Garthoff, L.H., L. Friedman, T.M. Farber, et al. 1977. Biochemical and cytogenetic effects in rats caused by short-term ingestion of Aroclor 1254 or Firemaster BP6. J. Toxicol. Environ. Health. 3: 769.

Garthoff, L.H., F.E. Cerra and E.M. Marks. 1981. Blood chemistry alterations in rats after single and multiple gavage administration of polychlorinated biphenyls. Toxicol. Appl. Pharmacol. 60(1): 33-44.

Gartner, L.M. and I.M. Arias. 1966. Studies of prolonged neonatal jaundice in the breast-fed infant. J. Pediat. 68(1): 54.

Gellert, R.J. 1978. Uterotrophic activity of PCB induction of precocious reproductive aging in neonatally treated female rats. Environ. Res. 16: 123.

Gellert, R.J. and C. Wilson. 1979. Reproductive function in rats exposed prenatally to pesticides and polychlorinated biphenyls. Environ. Res. 18: 437.

Ghiasuddin, S.M., R.E. Menzer and J.O. Nelson. 1976. Metabolism of 2,5,2'-trichloro-, 2,5,2',5'-tetrachloro-, and 2,4,5,2',5'-pentachlorobiphenyl in rat hepatic microsomal system. Toxicol. Appl. Pharmacol. 36: 187-194.

Gillette, J.R. 1967. Individually different responses to drugs according to age, sex, and functional or pathological state. In: Drug Responses in Man, G. Wolstenholme and R. Porter, Ed. Churchill, London. p. 28.

Goldstein, J.A., P. Hickman, H. Bergman, J.D. McKinney and M.P. Walker. 1977. Separation of pure polychlorinated biphenyls isomers into two types of inducers on the basis of induction of cytochrome P-450 or P-448. Chem. Biol. Interact. 17: 69.

Goto, M. and K. Higuchi. 1969. The symptomatology of Yusho (chlorobiphenyls poisoning) in dermatology. Fukuoka Acta. Med. 60: 409. (Jap.) (Cited in Kuratsune et al., 1972)

Goto, M., K. Sugiura, M. Hattori, T. Miyagawa and M. Okamura. 1974a. Metabolism of 2,3-dichlorobiphenyl-^{14}C and 2,4,6-trichlorobiphenyl-^{14}C in the rat. Chemosphere. 5: 227-232.

Goto, M., K. Sugiura, M. Hattori, T. Miyagawa and M. Okamura. 1974b. Metabolism of 2,3,5,6-tetrachlorobipheynl-^{14}C and 2,3,4,5,6-pentachlorobiphenyl-^{14}C in the rat. Chemosphere. 5: 233-238.

Goto, M., M. Hattori and K. Sugiura. 1975. Metabolism of pentachloro- and hexachlorobiphenyls in the rat. Chemosphere. 3: 177-180.

Gottlieb, A.A. and S.R. Waldman. 1972. The multiple functions of macrophages in immunity. In: Macrophages and Cellular Immunity, A. Laskin et al., Ed. CRC Press, West Palm Beach, FL. p. 13-44.

Grant, D.L. and W.E.J. Phillips. 1974. The effect of age and sex on the toxicity of Aroclor(R) 1254, a polychlorinated biphenyl, in the rat. Bull. Environ. Contam. Toxicol. 12: 145-152.

Green, S., J.V. Carr, K.A. Palmer and E.J. Oswald. 1975a. Lack of cytogenetic effects in bone marrow and spermatogonial cells in rats treated with polychlorinated biphenyls Aroclors 1242 and 1254. Bull. Environ. Contam. Toxicol. 13: 14.

Green, S., F.M. Sauro and L. Friedman. 1975b. Lack of dominant lethality in rats treated with polychlorinated biphenyls (Aroclors 1242 and 1254). Food Cosmet. Toxicol. 13: 507.

Hansell, M.M. and D.J. Ecobichon. 1974. Effects of chemically pure chlorobiphenyls on the morphology of rat liver. Toxicol. Appl. Pharmacol. 28: 418.

Hansen, L.G., C.S. Byerly, R.L. Metcalf and R.F. Bevill. 1975. Effect of PCB mixture on swine reproduction. Am. J. Vet. Res. 36: 23.

Haque, R., D.W. Schmedding and V.H. Freed. 1974. Aqueous solubility, adsorption and vapor behavior of polychlorinated biphenyl Aroclor 1254. Environ. Sci. Technol. 8(2): 139-142. (Cited in Callahan et al., 1979)

Hasegawa, T., M. Ogata and K. Tomokuni. 1982. Inhibition of oxidative phosphorylation of rat liver mitochondria by PCB and ferric chloride: A preliminary report. Ind. Health. 20(3): 273-275.

Hashimoto, K., S. Akasaka, Y. Takagi, et al. 1976. Distribution and excretion of [^{14}C]polychlorinated biphenyls after their prolonged administration to male rats. Toxicol. Appl. Pharmacol. 37: 415-423.

Hass, J.R., L.T. Jao, N.K Wilson and H.B. Matthews. 1977. Metabolism of 4-chlorobiphenyl and 4,4'-dichlorobiphenyl in the rat: Qualitative and quantitative aspects. J. Agric. Food Chem. 25: 1330.

Hausch, C., A. Vittoria, C. Silipo and P. Jow. 1974. Partition coefficients and the structure activities relationship of the anaesthetic gases. J. Med. Chem. 18(6): 546-548.

Heddle, J.A. and W.R. Bruce. 1977. Comparison of tests for mutagenicity or carcinogenicity using assays for sperm abnormalities, formation of micronuclei and mutations in Salmonella. In: Origins of Human Cancer, H.H. Hiatt et al., Ed. Cold Spring Harbor Conf. Cell Prolif., Cold Spring Harbor Lab., Cold Spring Harbor, NY. 4: 1549.

Hetling, L., E. Horn and J. Tofflemire. 1978. Hudson River PCB study results. NY State Dept. Environ. Conservation, Albany, NY. (unpublished) (Cited in Callahan et al., 1979)

Hinton, D.E., H. Glaumann and B.F. Trump. 1978. Studies on the cellular toxicity of polychlorinated biphenyls (PCBs). I. Effect of PCBs on microsomal enzymes and on synthesis and turnover of microsomal and cytoplasmic lipids of rat liver - a morphological and biochemical study. Verchoms Arch. B Cell Pathol. 27: 279-306.

Hirayama, C., M. Okumura, J. Nagai and Y. Masuda. 1974. Hypobilirubinemia in patients with polychlorinated biphenyls poisoning. Clin. Chim. Acta. 55: 97.

Hofvander, Y., U. Hagman, C.E. Linder, R. Vaz and S.A. Slorach. 1981. WHO collaborative breast feeding study. I. Organochlorine contaminants in individual samples of Swedish human milk, 1978-1979. Acta. Paediatr. Scand. 70(1): 3-8.

Hollifield, H.C. 1979. Rapid nephelometric estimate of water solubility of highly insoluble organic chemicals of environmental interest. Bull. Environ. Contam. Toxicol. 23: 579-586.

Hoopingarner, R., A. Samuel and D. Krause. 1972. Polychlorinated biphenyls interactions with tissue culture cells. Environ. Health Perspect. 1: 155-158.

Hori, S., H. Obana, K., T. Kashimoto, et al. 1982. Effect of polychlorinated biphenyls and polychlorinated quaterphenyls in cynomolgus monkey (Macaca fascicularis). Toxicology. 24(2): 123-139.

Horio, F., N. Kato and A. Yoshida. 1981. Interaction of PCB and cadmium for drug-metabolzing enzyme, plasma cholesterol and urinary ascorbic acid in rats. Nutr. Rep. Int. 24(3): 431-442.

Hsu, I.C., J.P. Van Miller, J.R. Allen. 1975a. Metabolic fate of ^{3}H-2,5,2',5'-tetrachlorobiphenyl in infant nonhuman primates. Bull. Environ. Contam. Toxicol. 14: 233.

Hsu, I.C., J.P. Van Miller, J.L. Seymour and J.R. Allen. 1975b. Urinary metabolites of 2,5,2',5'-tetrachlorobiphenyl in the non-human primate. Proc. Soc. Exp. Biol. Med. 150: 185.

Hutton, J.J., J. Meier and C. Hackney. 1979. Comparison of the in vitro mutagenicity and metabolism of dimethylnitrosamine and benzo[a]pyrene in tissues from inbred mice treated with phenobarbital, 3-methylcholanthrene or polychlorinated biphenyls. Mutat. Res. 66: 75.

Hutzinger, O., D.M. Nash, S. Safe, et al. 1972. Polychlorinated biphenyls: Metabolic behavior of pure isomers in pigeons, rats and brook trout. Science. 178: 312-318. (Cited in Callahan et al., 1979)

Hutzinger, O., W.D. Jamieson, S. Safe, L. Paulmann and R. Ammon. 1974. Identification of metabolic dechlorination of highly chlorinated biphenyl in rabbit. Nature (Lond.). 252: 698-699.

Hutzinger, O., S. Safe and V. Zitko. 1974. The Chemistry of PCBs. Chemical Rubber Publishing Co., Cleveland, OH.

IARC (International Agency for Research on Cancer). 1974. Polychlorinated Biphenyls. In: Some Anti-thyroid and Related Substances, Nitrofurans and Industrial Chemicals. IARC Monographs on the Evaluation of Carcinogenic Risk of Chemicals to Man. WHO, IARC, Lyon, France. Vol. 7, p. 261.

IARC (International Agency for Research on Cancer). 1978. Polychlorinated biphenyls. In: Polychlorinated Biphenyls and Polybrominated Biphenyls. IARC Monographs on the Evaluation of the Carcinogenesis Risk of Chemicals to Humans. WHO, IARC, Vol. 18, Lyon, France. p. 43.

Iatropoulos, M.J., G.R. Felt, H.P. Adams, et al. 1977. Chronic toxicity of 2,5,4'-trichlorobiphenyl in young rhesus monkeys. II. Histopathology. Toxicol. Appl. Pharmacol. 41(3): 629-638.

Iatropoulos, M.J., J. Bailey, H.P. Adams, F. Coulston and W. Hobson. 1978. Response of nursing infant rhesus to clophen A-30 or hexachlorobenzene given to their lactating mothers. Environ. Res. 16(1-3): 38-47.

Ito, N., H. Nagasaki, M. Arai, S. Makiura, S. Sugihara and K. Hirao. 1973. Histopathologic studies on liver tumorigenesis induced in mice by technical polychlorinated biphenyls and its promoting effect on liver tumors induced by benzene hexachloride. J. Natl. Cancer Inst. 51: 1637-1646.

Ito, N., H. Nagasaki, S. Makiura and M. Arai. 1974. Histopathological studies on liver tumorigenesis in rats treated with polychlorinated biphenyls. Gann. 65: 545-549.

Ito, N., M. Tatematsu, M. Hirose, K. Nakanishi and G. Murasaki. 1978. Enhancing effects of chemicals on production of hyperplastic liver nodules induced by N-2 fluorenyl-acetamide in hepatectomized rats. Gann. 69: 143.

Iverson, F., J. Truelove and S.L. Hierlihy. 1982. Hepatic microsomal enzyme induction by Aroclors 1246 and 1254 in cynomolgus monkeys. Food Chem. Toxicol. 20(3): 307-310.

Jacobson, J.L., G.G. Fein, S.W. Jacobson, et al. 1984. The transfer of polychlorinated biphenyls (PCBs) and polybrominated biphenyls (PBBs) across the human placenta and into maternal milk. Am. J. Public Health. 74: 378-379.

Jensen, S. and G. Sundstrom. 1974. Metabolic hydroxylation of a chlorobiphenyl containing only isolated unsubstituted positions -- 2,2'4,4',5,5'-hexachlorobiphenyl. Nature (Lond.) 251: 219-220.

Jones, J.W. and H.S. Alden. 1936. An acneform dermatergosis. Arch. Dermat. Syphilol. 33: 1022.

Jonsson, H.T. Jr., E.M. Walker, W.B. Green M.D. Hughson and G.R. Hennigar. 1981. Effect of prolonged exposure to dietary DDT and PCB on rat liver morphology. Arch. Environ. Contam. Toxicol. 10(2): 171-183.

Kaiser, K.L.E. and P.T.S. Wong. 1974. Bacterial degradation of PCBs: I. Identification of some metabolic products from Aroclor 1242. Bull. Environ. Contam. Toxicol. 11(2): 291-296. (Cited in Callahan et al., 1979)

Kasza, L., W.T. Collins, C.C. Capen, L.H. Garthoff and L. Friedman. 1978a. Comparative toxicity of polychlorinated biphenyls and polybrominated biphenyl in the rat thyroid gland: Light and electron microscopic alterations after subacute dietary exposure. J. Environ. Pathol Toxicol. May-June(5): 587-599.

Kasza, L., M.A. Weinberger, D.E. Hinton, et al. 1978b. Comparative toxicity of polychlorinated biphenyls and polybrominated biphenyl in the rat liver: Light and electron microscopic alterations after subacute dietary exposure. J. Environ. Pathol. Toxicol. Jan-Feb(3): 241-257.

Kato, N., K. Kawai and A. Yoshida. 1981. Effect of dietary level of ascorbic acid on the growth, hepatic lipid peroxidation, and serum lipids in guinea pigs fed polychlorinated biphenyls. J. Nutr. 111(10): 1727-1733.

Kerkvliet, N.I. and D.J. Kimeldorf. 1977a. Inhibition of tumor growth in rats by feeding a polychlorinated biphenyls, Aroclor 1254. Bull. Environ. Contam. Toxicol. 18: 243.

Kerkvliet, N.I. and D.J. Kimeldorf. 1977b. Antitumor activity of a polychlorinated biphenyls mixture, Aroclor 1254, in rats inoculated with Walker 256 carcinoma cells. J. Natl. Cancer Inst. 59: 951.

Khan, H.M. and L.K. Cutkomp. 1982. Effects of DDT, DDE and PCBs on mitochondrial respiration. Bull. Environ. Contam. Toxicol. 29(5): 577-585.

Kikuchi, M. and Y. Masuda. 1976. The pathology of Yusho. In: PCB Poisoning and Pollution, K. Higuchi, Ed. New York Academic Press. p. 69-86. (Cited in NIOSH, 1977)

Kikuchi, M., et al. 1969. An autopsy case of stillborn of chlorobiphenyls poisoning. Fukuoka Acta. Med. 60: 489. (Jap.) (Cited in Kuratsune et al., 1972)

Kimbrough, R.D. 1974. The toxicity of polychlorinated polycyclic compounds and related chemicals. CRC Crit. Rev. Toxicol. 2: 445.

Kimbrough, R.D. 1979. The carcinogenic and other chronic effects of persistent halogenated organic compounds. Ann. NY Acad. Sci. 320: 415.

Kimbrough, R.D. and R.E. Linder. 1974. Induction of adenofibrosis and hepatomas in the liver of Balb/cJ mice by polychlorinated biphenyls (Aroclor 1254). J. Natl. Cancer Inst. 53: 547.

Kimbrough, R.D., R.E. Linder and T.B. Gaines. 1972. Morphological changes in livers of rats fed polychlorinated biphenyls. Arch. Environ. Health. 25: 354.

Kimbrough, R.D., R.A. Squire, R.E. Linder, J.D. Strandberg, R.J. Montali and V.W. Burse. 1975. Induction of liver tumors in Sherman strain female rats by polychlorinated biphenyls Aroclor 1260. J. Natl. Cancer Inst. 55: 1453.

Kimbrough, R., J. Buckley, L. Fishbein, et al. 1978. Animal toxicity. Environ. Health Perspect. 24: 173.

Kimura, N.T. and T. Baba. 1973. Neoplastic changes in the rat liver induced by polychlorinated biphenyls. Gann. 64: 105.

Kintzman, R., R.M. Welch and A.H. Conney. 1967. Factors influencing steroid hydroxylases in liver microsomes. Adv. Enzyme Regulation. 4: 149-160.

Kling, D., J. Kunkle, A.S. Roller and W. Gambler. 1978. Polychlorinated biphenyls: _In vivo_ and _in vitro_ modifications of cholesterol and fatty acid biosynthesis. J. Environ. Pathol. Toxicol. 1(6): 813.

Koch, R.B., D. Desaiah, H.H. Yap and L.K. Cutkomp. 1972. Polychlorinated biphenyls: Effect of long-term exposure on ATPase activity in fish, _Pimephales promelas_. Bull. Environ. Contam. Toxicol. 7: 87-92.

Koller, L.D. 1977. Enhanced polychlorinated biphenyls lesions in Moloney leukemia virus-infected mice. Clin. Toxicol. 11(1): 107-116.

Koller, L.D. and J.G. Zinkl. 1973. Pathology of polychlorinated biphenyls in rabbits. Am. J. Pathol. 70: 363.

Komives, G.K. 1979. Body weights, food intakes, and water intakes in rats during daily administration of closely controlled doses of polychlorinated biphenyls. Bull. Environ. Contam. Toxicol. 22(6): 761-766.

Komives, G.K. and G. Alayoku. 1980. Body temperatures and weights in rats during daily administration of closely controlled doses of polychlorinated biphenyls. Bull. Environ. Contam. Toxicol. 25(6): 913-917.

Kraul, I. and O. Karlog. 1976. Persistent organochlorinated compounds in human organs collected in Denmark 1972-73. Acta. Pharmacol. Toxicol. (Kbh). 38(1): 38-73.

Kreiss, K., M.M. Zack, R.D. Kimbrough, L.L. Needham, A.L. Smrek and B.T. Jones. 1981. Association of blood pressure and polychlorinated biphenyls levels. J. Am. Med. Assoc. 245(24): 2505-2509.

Kurachi, M. 1983. Studies on excretion and accumulation of PCBs in connection with their partial metabolism in the animal body. Part II. A new sulfur-containing derivative and possibility of conjugate formation of PCBs in mice or rats. Agric. Biol. Chem. 47(6): 1183-1191.

Kurachi, M. and T. Mio. 1983a. Studies on excretion and accumulation of PCBs in connection with their partial metabolism in the animal body. Part III. On the formation of a conjugate of PCBs with glutathione and its further metabolism in mice or rats. Agric. Biol. Chem. 47(6): 1193-1199.

Kurachi, M. and T. Mio. 1983b. Studies on excretion and accumulation of PCBs in connection with their partial metabolism in the animal body. Part I. On fluctuation of PCBs under various unnatural conditions in mice. Agric. Biol. Chem. 47(6): 1173-1181.

Kuratsune, M., et al. 1969. An epidemiologic study on "Yusho" or chlorobiphenyls poisoning. Fukouka Acta. Med. 60: 513. (Jap.) (Cited in Kuratsune et al., 1972)

Kuratsune, M., T. Yoshimura, J. Matsuzaka and A. Yamaguchi. 1972. Epidemiologic study on Yusho, a poisoning caused by ingestion of rice oil contaminated with a commercial brand of polychlorinated biphenyls. Environ. Health Perspect. 1: 119-128.

Kuwabara, K., T. Yakushiji, I. Wantanabe, S. Yoshida, K. Kyoama and N. Kunita. 1979. Levels of polychlorinated biphenyls in blood of breast-fed children whose mothers are non-occupationally exposed to PCBs. Bull. Environ. Contam. Toxicol. 21(4-5): 458-462.

Lake, B.G., M.A. Collins, R.A. Harris and S.D. Gangolli. 1979. The induction of hepatic and extrahepatic xenobiotic metabolism in the rat and ferret by a polychlorinated biphenyls mixture (Aroclor 1254). Xenobiotica. 9: 723.

Lawrence, C. 1977. PCB? and adenoma. New Engl. J. Med. 296: 108.

Lester, R. and R. Schmid. 1964. Bilirubin metabolism. New Engl. J. Med. 270(15): 779.

Lewin, C., W.A. McBlain and F.H. Wolfe. 1972. Acute intraperitoneal toxicity of DDT and PCBs in mice using two solvents. Bull. Environ. Contam. Toxicol. 8: 245-250.

Lin, F.S., M.T. Hsia and J.R. Allen. 1979. Acute hepatotoxicity of a tetrachloribiphenyl-changes in the hepatocyte ultrastructure and plasma membrane-bound enzymes. Arch. Environ. Contam. Toxicol. 8(3): 321-333.

Linder, R.E., T.B. Gaines and R.D. Kimbrough. 1974. The effect of PCB on rat reproduction. Food Cosmet. Toxicol. 12: 63.

Litterst, C.L., T.M. Farber, A.M. Baker and E.J. Van Loon. 1972. Effect of polychlorinated biphenyls on hepatic microsomal enzymes in the rat. Toxicol. Appl. Pharmacol. 23: 112.

Lokietz, H., R.M. Dowben and D.Y. Hsia. 1963. Studies on the effect of Novobiocin and Glucuronyl transferase. Pediatrics. 32: 47.

Loose, L.D., K.A. Pittman, K.-F. Benitz, J.B. Sitknorth, W. Mueller and F. Coulston. 1978a. Environmental chemical-induced immune dysfunction. Ecotoxicol. Environ. Safety. 2: 173.

Loose, L.D., J.B. Silkworth, K.A. Pittman, K.-F. Benitz and W. Mueller. 1978b. Impaired host resistance to endotoxic and malaria in polychlorinated biphenyl- and hexachlorobenzene-treated mice. Inf. Immun. 20(1): 30.

Lutz, R.J., R.L. Dedrick, H.B. Matthews, T.E. Eling and M.W. Anderson. 1977. A preliminary pharmacokinetic model for several chlorinated biphenyls in the rat. Drug Metab. Dispos. 5: 386.

Mabey, W.R., J.H. Smith, R.T. Podoll, et al. 1981. Aquatic fate process data for organic priority pollutants. U.S. EPA, Washington, DC. EPA 440/4-81-014.

Mackay, D. 1982. Correlation of bioconcentration factors. Environ. Sci. Technol. 16: 274-278.

Mackay, D. and P.J. Leinonen. 1975. Rate of evaporation of low-solubility contaminants from water bodies to atmosphere. Environ. Sci. Technol. 9(13): 1178-1180.

Makiura, S., H. Aoe, Seiichi, K. Hiaro, M. Arai and N. Ito. 1974. Inhibitory effect of polychlorinated biphenyls on liver tumorigenesis in rats treated with 3'-methyl-4-dimethylaminobenzene, N-2-fluorenylacetamide and diethylnitrosamine. J. Natl. Cancer Inst. 53: 1253.

Marks, T.A., G.L. Kimmel and R.E. Staples. 1981. Influence of symmetrical polychlorinated biphenyls isomers on embryo and fetal development in mice. I. Teratogenicity of 3,3'4,4'5,5'-hexachlorobiphenyl. Toxicol. Appl. Pharmacol. 61(2): 269-276.

Maroni, M., A. Colombi, G. Arbosti, S. Cantoni and V. Foa. 1981. Occupational exposure to polychlorinated biphenyls in electrical workers. II. Health effects. Br. J. Ind. Med. 38(1): 55-60.

Masuda, Y, R. Kagawa and M. Kuratsune. 1974. Comparison of polychlorinated biphenyls in Yusho patients and ordinary persons. Bull. Environ. Contam. Toxicol. 11: 213-216.

Masuda, Y., R. Kagawa and H. Kuroki. 1978. Transfer of polychlorinated biphenyls to the foetuses and offspring of mice. Food Comset. Toxicol. 16: 33.

Masuda, Y., R. Kagawa, H. Kuroki, S. Tokudome and M. Kuratsune. 1979. Transfer of various polychlorinated biphenyls to the foetuses and offspring of mice. Food Cosmet. Toxcol. 17(6): 623-627.

Matthews, H.B. and M.W. Anderson. 1975a. The distribution and excretion of 2,4,5,',5'-pentachlorobiphenyl in the rat. Drug Metab. Dispos. 3: 211.

Matthews, H.B. and M.W. Anderson. 1975b. Effect of chlorination on the distribution and excretion of polychlorinated biphenyls. Drug. Metab. Dispos. 3: 371.

Mattsson, R., A. Mattsson, J.E. Kihlstroem and K. Lingahl-Kiessling. 1981. Effect of hexachlorinated biphenyl on lymphoid organs and resorption of fetuses in pregnant mice. Arch. Environ. Contam. Toxicol. 10(3): 281-288.

McConnell, E.E. and J.D. Kinney. 1978. Exquisite toxicity in the guinea pig to structurally similar halogenated dioxins, furans, biphenyls and naphthalenes. Toxicol. Appl. Pharmacol. 45: 298.

McConnell, E.E. 1980. Halogenated biphenyls, terphenyls, naphthalenes, dibenzodioxins and related products. Acute and chronic toxicity, carcinogenesis, reproduction, teratogenesis and mutagenesis in animals. Toxicol. Environ. Health. 4: 109-150.

McCormach, K.M., P. Melrose, D.E. Rickert, J.G. Dent, J.E. Gibson and J.B. Hook. 1979. Concomitant dietary exposure to polychlorinated biphenyls and polybrominated biphenyls: Tissue distribution and arylhydrocarbon hydroxylase activity in lactating rats. Toxicol. Appl. Pharmacol. 47: 95.

McNulty, W.P., G.M. Becker and H.T. Cory. 1980. Chronic toxicity of 3,3'4,4'- and 2,2'5,5'-tetrachlorobiphenyls in rhesus macques. Toxicol. Appl. Pharmacol. 56(2): 182-190.

Meigs, J.W., J.J. Albom and B.L. Kartin. 1954. Chloracne from an unusual exposure to Aroclor. J. Am. Med. Assoc. 154: 1417.

Mes, J. and D.J. Davies. 1979. Presence of polychlorinated biphenyls and organochlorine pesticide residues and the absence of polychlorinated terphenyls in Canadian human milk samples. Bull. Environ. Contam. Toxicol. 21(3): 381-387.

Mes, J., D.W. Campbell, R.N. Robinson and D.J.A. Davies. 1977. Polychlorinated biphenyls and organochlorine pesticides in adipose tissue of Canadians. Bull. Environ. Contam. Toxicol. 17(2): 196-203.

Miller, J.W. 1944. Pathologic changes in animals exposed to a commercial chlorinated diphenyl. Pub. Health Rep. 59: 1085.

Miller, R.W. 1977. Pollutants in breast milk: PCBs and cola-colored babies (editorial). J. Pediatr. 90(3): 510-511.

Miniats, O.P., N.S. Platonow and H.D. Geissinger. 1978. Experimental polychlorinated biphenyl toxicosis in germfree pigs. Can. J. Comp. Med. 42: 192-199.

Mizunoya, Y., S. Taniguchi, K. Kusumoto, et al. 1974. Effects of polychlorinated biphenyls on fetuses and offsprings in rats. J. Food Hyg. Soc. (Jap.) 15: 252.

Mizutani, T., K. Hidaka and M. Matsumoto. 1977. A comparative study on accumulation and elimination of tetrachlorobiphenyl isomers in mice. Bull. Environ. Contam. Toxicol. 18: 452.

Moein, G.J., A.J. Smith, Jr. and P.L. Stewart. 1976. Follow-up study of the distribution and fate of PCBs and benzenes in soil and ground water samples after an accidental spill of transformer fluid. In: Proc. of 1976 Natl. Conf. on Control of Hazardous Material Spills. Information Transfer, Inc., Rockville, MD. p. 368-372. (Cited in Callahan et al, 1979)

Monsanto Industrial Chemical Corp. 1974. BCB's Aroclors Tech. Bull. O/PC 306A, St. Louis, MO.

Morgan, R.W., J.M. Ward and P.E. Hartman. 1981. Aroclor 1254-induced intestinal metaplasia and adenocarcinoma in the glandular stomach of F344 rats. Cancer Res. 41(12): 5052-5059.

Morrison, R.T. and R.N. Boyd. 1973. Organic Chemistry, 3rd ed. Allyn and Bacon, Boston, MA. 1258 p.

Muehlebach, S. and M.H. Bickel. 1981. Pharmacokinetics in rats of 2,4,5,2',4',5'-hexachlorobiphenyl, an unmetabolizable lipophilic model compound. Xenobiotica. 11(4): 249-257.

Nagasaki, H., S. Tomii, T. Mega, M. Marugami and N. Ito. 1972. Hepatocarcinogenicity of polychlorinated biphenyls in mice. Gann. 63: 805.

Nagayama, J., M. Kurastune and Y. Masuda. 1976. Determination of chlorinated dibenzofurans in Kanechlors and "Yusho oil". Bull. Environ. Contam. Toxicol. 15: 9-13.

Narbonne, J.F. 1979. Effects of age and sex on liver responses of phenoclor DP6, a polychlorinated biphenyls in the rat. Bull. Environ. Contam. Toxicol. 22(1): 49-54.

NAS (National Academy of Science). 1977. Drinking Water and Health. National Academy Press, Washington, DC.

NAS (National Academy of Science). 1980. Drinking Water and Health, Vol. 3. National Academy Press, Washington, DC. p. 133.

NCI (National Cancer Institute). 1975. Third National Cancer Survey, Bethesda, MD. Incidence Data, March, 1975. (Cited in Bahn et al., 1976)

NCI (National Cancer Institute). 1978. Bioassay of Aroclor (trademark) 1254 for possible carcinogenicity. CAS No. 27323-18-8. NCI Carcinogenesis Tech. Rep. Ser. No. 38.

Nelson, N., B. Hammon, C.T. Nisbet, A.F. Sarafirm and W.H. Drwey. 1972. Polychlorinated biphenyls - environmental impact. Environ. Res. 5: 249.

Nilsson, B. and C. Ramel. 1974. Genetic tests on *Drosophila melanogaster* with polychlorinated biphenyls (PCB). Hereditas. 77: 319-322.

NIOSH (National Institute for Occupational Safety and Health). 1977. Criteria for a Recommended Standard...Occupational Exposure to Polychlorinated Biphenyls (PCBs). U.S. DHEW, PHS, CDC, Rockville, MD.

Nishihara, Y. 1983. Effects of polychlorinated biphenyls (Kanechlor-400) on isolated rat liver mitochondria. Arch. Environ. Contam. Toxicol. 12(5): 527-522.

Nishizumi, M. 1970. Light and electron microscope study of chlorobiphenyl poisoning in mouse and monkey liver. Arch. Environ. Health. 21: 620.

Nishizumi, M. 1976. Radioautographic evidence for adsorption of polychlorinated biphenyls through the skin. Ind. Health. 14: 41-44.

Nishizumi, M. 1980. Reduction of diethylnitrosamine-induced hepatoma in rats exposed to polychlorinated biphenyls through their dams. Gann. 71(6): 910-912.

Norback, D.H., J.L. Seymour, K.M. Knieriem, R.E. Peterson and J.R. Allen. 1976. Biliary metabolites of 2,5,2',5'-tetrachlorobiphenyl in the rat. Res. Commun. Chem. Pathol. Pharmacol. 14: 527-533.

Norback, D.H., E. Mack, K.A. Blomquist and J.R. Allen. 1978. Metabolic study of 2,4,5,2',4',5'-hexachlorobiphenyl in rhesus monkeys. Toxicol. Appl. Pharmacol. 45: 331.

Nyhan, W.L. 1961. Toxicity of drugs in the neonatal period. J. Pediat. 59(1): 1.

Oesterle, D. and E. Deml. 1983. Promoting effect of polychlorinated biphenyls on development of enzyme-altered islands in livers of weanling and adult rats. J. Cancer Res. Clin. Oncol. 105(2): 141-146.

Ohnishi, Y. and T. Kohmo. 1979. Polychlorinated biphenyls poisoning in monkey eye. Incvest. Ophthahmol. Visual Sci. 18: 981-984.

Oishi, S., M. Morita and H. Fekuda. 1978. Comparative toxicity of polychlorinated biphenyls and dibenzofurans in rats. Toxicol. Appl. Pharmacol. 43(1): 13-22.

Oloffs, P.C., L.J. Albright and S.Y. Szeto. 1972. Fate and behavior of five chlorinated hydrocarbons in three natural waters. Can. J. Microbiol. 18: 1393-1398. (Cited in Callahan et al., 1979)

Oloffs, P.C., L.S. Albright, S.Y. Szeto and J. Jau. 1973. Factors affecting the behavior of five chlorinated hydrocarbons and their sediments. J. Fish. Res. Board Can. 30: 1619. (Cited in Callahan et al., 1979)

Orberg, J. 1977. Placental and mammary transfer of two PCBs (2,4',5-TCB and 2,2',4,4',5,5'-HCB) and their effect on reproductive capacity in mice. Ambio. 6: 228.

Orberg, J. 1978. Effects of pure chlorobiphenyls (2,4',5-trichlorobiphenyl and 2,2',4,4'5,5'-hexachlorobiphenyl) on the reproductive capacity in female mice. Acta. Pharmacol. Toxicol. 42: 323.

Orberg, J. and C. Ingvast. 1977. Effects of pure chlorobiphenyls (2,4',5-trichlorobiphenyl and 2,2'4,4'5,5'-hexachlorobiphenyl) on the disappearance of ^{14}C from the blood plasma after intravenous injection of 4-^{14}C-progesterone and on the hepatic drug metabolizing system in the female rat. Acta. Pharmacol. Toxicol. 41: 11.

Orberg, J. and J.E. Xihlstrom. 1973. Effects of long-term feeding of polychlorinated biphenyls (PCB, Clophen A60) on the length of the oestrous cycle and on the frequency of implanted ova in the mouse. Environ. Res. 6: 176.

Ouw, H.K., G.R. Simpson and D.S. Siyali. 1976. Use and health effects of Aroclor 1242, a polychlorinated biphenyls in an electrical industry. Arch. Environ. Health. 31: 189.

Pardini, R.S. 1971. Polychlorinated biphenyls (PCB). Effect on mitochondrial enzyme system. Bull. Environ. Contam. Toxicol. 6: 539-545.

Paris, D.F., W.C. Steen and G.L. Baughman. 1978. Role of the physicochemical properties of Aroclor 1016 and 1242 in determining their fate and transport in aquatic environments. Chemosphere. 7(4): 319-325. (Cited in Callahan et al., 1979)

Parkinson, A., P.E. Thomas, D.E. Reik, et al. 1983. Differential time course of induction of rat liver microsomal cytochrome P 450 siozymes and epoxide hydrolase by Aroclor 1254. Arch. Biochem. Biophys. 225(1): 203-215.

Parkki, M.G., J. Marniemi and H. Vainio. 1977. Long-term effects of single and combined doses of DDT and PCB on drug-metabolizing exzymes in rat liver. J. Toxicol. Environ. Health. 3(5-6): 903-911.

Peakall, D.B. 1972. Polychlorinated biphenyls: Occurrence and biological effects. Residue Rev. 44: 1.

Peakall, D.B. 1975. PCBs and their environmental effects. CRC Critical Rev. Environ. Control. 5: 469-508.

Peakall, D.B., J.L. Lincer and S.E. Bloom. 1972. Embryonic mortality and chromosomal alterations caused by Aroclor 1254 in ring doves. Environ. Health Perspect. 1: 103-104.

Pereira, M.A., S.L. Herren, A.L. Britt and M.M. Khoury. 1982. Promotion by polychlorinated biphenyls of enzyme-altered foci in rat liver. Cancer Lett. 15(2): 185-190.

Pittman, K.A., K.F. Benitz and W. Mueller. 1979. Modification of the immune response by organochlorine xenobiotics. Drug Chem. Toxicol. 2(1-2): 111-132.

Pomerantz, I., J. Burke, D. Firestone, J. McKinney, J. Roach and W. Trotter. 1978. Chemistry of PCBs and PBBs. Environ. Health Perspect. 24: 133-146.

Ramanathan, M. 1984. Water pollution. In: Kirk-Othmer Encyclopedia of Chemical Technology, 3rd ed., M. Grayson and D. Eckroth, Ed. John Wiley and Sons, Inc., New York. Vol. 24, p. 306.

Reuber, M.D. 1968. Histogenesis of cholangiofbrosis and well differentiated cholangiocarcinoma in Syrian hamsters given 2-acetoamidofluorene or 2-diacetoamidofluorene. Gann. 59: 239.

Ringer, R.K., R.J. Aulerich and M. Zabik. 1972. Effect of dietary polychlorinated biphenyls on growth and reproduction of mink. 164th Natl. Meet. Am. Chem. Soc. 12: 149.

Risebrough, R.W., P. Reiche, D.B. Peakall, S.G. Herman and M.N. Kirven. 1968. Polychlorinated biphenyls in the global ecosystem. Nature. 220: 1098.

Roach, J.A.G. and I.H. Pomerantz. 1974. The finding of chlorinated dibenzofurans in a Japanese polychlorinated biphenyl sample. Bull. Environ. Contam. Toxicol. 12: 338-342.

Rogan, W.J., A. Bogniewska and T. Damstra. 1980. Pollutants in breast milk. N. Engl. J. Med. 302: 1450.

Rosin, D.L. and B.R. Martin. 1981. Neurochemical and behavioral effects of polychlorinated biphenyls in mice. Nurotoxicology. 2: 749-764.

Rosin, D.L. and B.R. Martin. 1983. Comparison of the effects of acute and subchronic administration of arochlor 1254, a commmercial mixture of polychlorinated biphenyls on pentobarbital-induced sleep time and [^{14}C]pentobarbital disposition in mice. J. Toxicol. Environ. Health. 11: 917-931.

Rozanova, L.F. 1943. [Toxicity of some chlorinated aromatic hydrocarbons]. Farmakol Toksikol. 6: 48. (Cited in NIOSH, 1977)

Safe, S. 1980. Halogenated biphenyls, terphenyls, napthalenes, dibenzodioxins and related products. Metabolism, uptake, storage and bioaccumulation. Toxicol. Environ. Health. 4: 81-107.

Safe, S., O. Hutzinger and D. Jones. 1975a. The mechanism of chlorobiphenyl metabolism. J. Agric. Food Chem. 23: 851-853.

Safe, S., N. Platonom and O. Hutzinger. 1975b. Metabolism of chlorobiphenyls in the goat and cow. J. Agric. Food Chem. 23: 259-261.

Safe, S., D. Jones and O. Hutzinger. 1976b. Metabolism of 4,4'-dihalogenobiphenyls. J. Chem. Soc. Perkin Trans. 1: 357.

Sager, D.B. 1983. Effect of postnatal exposure to polychlorinated biphenyls on adult male reproductive function. Environ. Res. 31(1): 76-94.

Sanders, O.T., R.L. Zepp and R.L. Kirkpatrick. 1974. Effect of PCB ingestion on sleeping times, organ weights, food consumption, serum corticosterone and survival of albino mice. Bull. Environ. Contam. Toxicol. 12(4): 394-399.

Schoeny, R. 1982. Mutagenicity testing of chlorinated biphenyls and chlorinated dibenzofurans. Mutat. Res. 101: 45.

Schoeny, R.S., C.C. Smith and J.C. Loper. 1979. Non-mutagenicity for Salmonella of the chlorinated hydrocarbons Aroclor 1254, 1,2,4-trichlorobenzene, mirex, and kepone. Mutat. Res. 68: 125.

Schwartz, L. 1943. An outbreak of halowax acne ('cable rash') among electricians. J. Am. Med. Assoc. 122: 158-161.

Shigematsu, N., S. Ishimaru and R. Saito, et al. 1978. Respiratory involvement in polychlorinated biphenyls poisoning. Environ. Res. 16(1-3): 92-100.

Shimada, T. and R. Sato. 1980. Covalent binding of polychlorinated biphenyls to rat liver microsomes _in vitro_: Nature of reactive metabolites and target macromolecules. Toxicol. Appl. Pharmacol. 55(3): 490-500.

Shull, L.R., M.R. Bleavins, B.A. Olson and R.J. Aulerich. 1982. Polychlorinated biphenyls (Aroclors 1016 and 1242): Effect on hepatic microsomal mixed-function oxidases in mink and ferrets. Arch. Environ. Contam. Toxicol. 11(3): 313-321.

Silbergeld, E.K. 1983. Health effects on PCBs: Occupational exposures. _In_: Proceedings PCB - Seminar, Scheveningen, The Hague, The Netherlands, September 28-30. p. 136-151.

Silkworth, J.B. and L.D. Loose. 1978. Cell mediated immunity in mice fed either Aroclor 1016 or hexachlorobenzene. Toxicol. Appl. Pharmacol. 45: 326.

Sipes, I.G. and B.R. Brown. 1976. An animal model of hepatotoxicity associated with halothane anesthesia. Anesthesiology. 45(6): 622-628.

Sipes, I.G., G.E. McLain, Jr., T.L. Podolsky and B.R. Brown, Jr. 1978. Bioactivation of halothane: Correlation with hepatotoxicity. Int. Congr. Serx-Excerpta Med. 440: 238.

Sivalingan, P.M., T. Yoshida and Y. Inada. 1973. Modes of inhibitory effects of PCBs on oxidative phosphorylation of mitochondria. Bull. Environ. Contam. Toxicol. 10: 242.

Smith, A.J., Jr., G.J. Moein and P.L. Stewart. 1976. Follow-up study of the distribution and fate of polychlorinated biphenyls and benzenes in soil and groundwater samples after an accidental spill of transformer fluid. NTIS PB-273984. p. 19.

Spencer, F. 1982. An assessment of the reproductive toxic potential of Aroclor 1254 in female Sprague-Dawley rats. Bull. Environ. Contam. Toxicol. 28(3): 290-297.

Stadnicki, S.S. and J.R. Allen. 1979. Toxicity of 2,2',5,5'-tetrachlorobiphenyl and its metabolites, 2,2',5,5'-tetrachlorobiphenyl-3,4-oxide and 2,2',5,5'-tetrachlorobiphenyl-4-of to cultured cells *in vitro*. Bull. Environ. Contam. Toxicol. 23(6): 788-796.

Street, J.C. and R.P. Sharma. 1975. Alteration of induced cellular and humoral immune responses by pesticides and chemicals of environmental concern: Quantitative studies of immunosuppression by DDT, Aroclor 1254, carbaryl, carbofuran, and methyl parathion. Toxicol. Appl. Pharmacol. 32: 587.

Street, J.C., F.M. Urry, D.J. Wagstaff and A.D. Blau. 1969. Comparative effects of polychlorinated biphenyls and organochlorine pesticides in induction of hepatic microsomal enzymes. Paper presented at 158th Natl. Meet. Am. Chem. Soc., New York.

Sundstrom, G., O. Hutzinger and S. Safe. 1976a. The metabolism of chlorobiphenyls - A review. Chemosphere. 5: 267.

Sundstrom, G., O. Hutzinger and S. Safe. 1976b. The metabolism of 2,2',4,4',5,5'-hexachlorobiphenyl by rabbits, rats and mice. Chemosphere. 4: 249.

Suzuki, T. 1980. Additive effects of dietary cadmium and PCB in rats. Agric. Biol. Chem. 44(9): 2209-2210.

Tabak, H.H., S.A. Quave, C.I. Mashni and E.F. Barth. 1981a. Biodegradability studies with organic priority pollutant compounds. J. Water Pollut. Control Fed. 53: 1503-1518.

Tabak, J.H., Quave, S.A., Mashni, C.I. and E.F. Barth. 1981b. Biodegradability studies for predicting the environmental fate of organic priority pollutants. In: Test Protocols for Environmentat Fate and Movement of Toxicants, Proc. of a Sym. Assoc. of Official Anal. Chem., 94th Ann. Mtg., Washington, DC. p. 267-328.

Taki, I., S. Hisanaga and Y. Amagase. 1969. Report on Yusho (chlorobiphenyls poisoning) pregnant women and their fetuses. Fukuoka Acta. Med. 60: 471. (Jap.) (Cited in Kuratsune et al., 1972)

Tanimura, T., M. Ema and T. Kihara. 1980. Effects of combined treatment with methylmercury and polychlorinated biphenyls (PCBs) on the development of mouse offspring. Sch. Med. 4: 163-198.

Tatematsu, M., K. Nakanishi, G. Murasaki, Y. Miyata, M. Hirose and N. Ito. 1979. Enhancing effect of inducers of liver microsomal enzymes on induction of hyperplastic liver nodules by N-2-fluorenylacetamide in rats. J. Natl. Cancer Inst. 63(6): 1411-1416.

Thomas, P.T. and R.D. Hinsdill. 1978. Effect of polychlorinated biphenyls on the immune responses of rhesus monkeys and mice. Toxicol. Appl. Pharmacol. 44: 41-51.

Torok, P. 1978. Delayed implantation and early developmental defects in the mouse caused by PCB - 2,2'-dichlorobiphenyl. Arch. Toxicol. 40: 249.

Torok, P. and L.W.D. Weber. 1981. Distribution of polychlorobiphenyls in pregnant mice and fetuses on various days of gestation. Arch. Environ. Contam. Toxicol. 10: 289.

Truelove, J., D. Grant, J. Mes, H. Tryphonas, L. Tryphonas and Z. Zawidzka. 1982. Polychlorinated biphenyl toxicity in the pregnant cynomolgus monkey: A pilot study. Arch. Environ. Contam. Toxicol. 11(5): 583-588.

Tucker, E.S., V.W. Saeger and O. Hicks. 1975. Activated sludge primary biodegradation of polychlorinated biphenyls. Bull. Environ. Contam. Toxicol. 14(6): 705-713. (Cited in Callahan et al., 1979)

Tuey, D.B. and H.B. Matthews. 1977. Pharmacokinetics of 3,3',5,5'-tetrachlorobiphenyl in the male rat. Drug Metab. Disp. 5: 444.

Tulp, M. Th.M. and O. Hutzinger. 1978. Some thoughts on aqueous solubilities and partition coefficients of PCB and the mathematical correlations between bioaccumulation and physicochemical properties. Chemosphere. 7(10): 849-860.

Uchiyama, M. T. Chiba and K. Noda. 1974. Co-carcinogenic effect of DDT and PCB feedings on methylcholanthrene-induced chemical carcinogenesis. Bull. Environ. Contam. Toxicol. 12: 687.

Urabe, H., M. Koda and M. Asahi. 1979. Present state of Yusho patients. NY Acad. Sci. Ann. 320: 273.

U.S. EPA. 1980a. Ambient Water Quality Criteria Document for PCBs. Environmental Criteria and Assessment Office, Cincinnati, OH. EPA 440/5-80-068.

U.S. EPA. 1980b. Guidelines and methodology used in the preparation of health effects assessment chapters of the consent decree water quality criteria. Federal Register. 45: 79347-79357.

Van Miller, J.P., I.C. Hsu and J.R. Allen. 1975. Distribution and metabolism of ^{3}H-2,5,2',5'-tetrachlorobiphenyl in rats. Proc. Soc. Exp. Biol. Med. 148: 682.

Villeneuve, D.C., B.L. Grant, K. Khera, D.J. Clegg, H. Baer and W.E.J. Phillips. 1971a. The fetotoxicity of a polychlorinated biphenyl mixture (Aroclor 1254) in the rabbit and in the rat. Environ. Physiol. 1: 67.

Villeneuve, D.C., D.L. Grant, W.E.J. Phillips, M.L. Clark and D.J. Clegg. 1971b. Effects of PCB administration on microsomal enzyme activity in pregnant rabbits. Bull. Environ. Contam. Toxicol. 6: 120-128.

Villeneuve, D.C., D.L. Grant and W.E.J. Phillips. 1972. Modification of pentobarbitol sleeping times in rats following chronic PCB ingestion. Bull. Environ. Contam. Toxicol. 7: 264.

Vos, J.G. and R.B. Beems. 1971. Dermal toxicity studies of technical polychlorinated biphenyls and fractions thereof in rabbits. Toxicol. Appl. Pharmacol. 19: 617.

Vos, J.G. and T. DeRoij. 1972. Immunosupressive activity of a polychlorinated biphenyl preparation on the hmoral immune response in guinea pigs. Toxicol. Appl. Pharmacol. 21: 549.

Vos, J.G. and J.H. Koeman. 1970. Comparative toxicologic study with polychlorinated biphenyls in chickens with special reference to porphyria, edema formation, liver necrosis and tissue residues. Toxicol. Appl. Pharmacol. 17: 656-668.

Vos, J.G. and E. Notenboom-Ram. 1972. Comparative toxicity study of 2,4,5,2',4',5'-hexachlorobiphenyl and a polychlorobiphenyl mixture in rabbits. Toxicol. Appl. Pharmacol. 23: 563.

Vos, J.G. and H. van Genderen. 1973. Toxicological aspects of immunosuppression. In: Pesticides in the Environment. A Continuing Controversy, W.B. Deichman, Ed. 8th Int. Conf. Toxicol. Occup. Med., Miami. Intercontinental Medical Book Co., NY.

Warshaw, R., A. Fischbein, J. Thornton, A. Miller and I.J. Selikoff. 1979. Decrease in vital capacity in PCB-exposed workers in a capacitor manufacturing facility. NY Acad. Sci. Ann. 320:(May) 277.

Wasserman, D., M. Wasserman, S. Circos and M. Djavaherian. 1973. Function of adrenal gland-zona fasciculata in rats receiving polychlorinated biphenyls. Environ. Res. 6: 334-338.

Wasserman, M., D.P. Nogueira, S. Cucos, et al. 1978. Organochlorine compounds in neoplastic and adjacent apparently normal gastric mucosa. Bull. Environ. Contam. Toxicol. 204(4):544-553.

Wasserman, M., M. Ron, B. Bercovici, D. Wassermann, S. Cucos and A. Pines. 1982. Premature delivery and organochlorine compounds: Polychlorinated biphenyls and some organochlorine insecticides. Environ. Res. 28(1): 106-112.

Watanabe, M. and T. Sugahara. 1981. Experimental formation of cleft palate in mice with polychlorinated biphenyls (PCB). Toxicology. 19(1): 49-53.

Wickizer, T.M., L.B. Brilliant, R. Copeland and R. Tilden. 1981. Polychlorinated biphenyl contamination of nursing mothers' milk in Michigan (USA). Am. J. Public Health. 71(2): 132-137.

Wolden, R., G.W. Lucier and C.M. Schiller. 1982. Effects of polychlorinated biphenyls on the development of intestinal and serum marker enzymes. J. Toxicol. Environ. Health. 9: 1-12.

Wolff, M.S. 1983. Occupationally derived chemicals in breast milk. Am. J. Ind. Med. 4: 259-281.

Wolff, T. and S. Hesse. 1977. Species difference of mixed-function oxidase induction between rabbits and rats afer pretreatment with polychlorinated biphenyls (PCBs). Biochem. Pharmacol. 26: 783-787.

Wong, P.T.S. and K.L.E. Kaiser. 1976. Bacterial degradation of PCBs: II. Rate studies. Bull. Environ. Contam. Toxicol. 13(2): 249-255. (Cited in Callahan et al., 1979)

Wyndham, C. and S. Safe. 1978. In vitro metabolism of 4-chlorobiphenyl by control and induced rat liver microsomes. Biochemistry. 17: 208.

Wyndham, C., J. Devenish and S. Safe. 1976. The in vitro metabolism, macromolecular binding and bacterial mutagenicity of 4-chlorobiphenyl, a model PCB substrate. Res. Commun. Chem. Pathol. Pharmacol. 15: 563.

Yagi, N., M. Kimura and Y. Itokawa. 1976. Sodium, potassium, magnesium, and calcium levels in polychlorinated biphenyl (PCB) poisoned rats. Bull. Environ. Contam. Toxicol. 16(5): 516-519.

Yalkowsky, S.H., S.C.L. Valvani and D. Mackay. 1983. Estimation of the aqueous solubility of some aromatic compounds. Residue Rev. 85: 43-55.

Yamaguchi, A., T. Yoshimura and M. Kuratsune. 1971. A survey on pregnant women having consumed mice oil contaminated with chlorobiphenyls and their babies. Fukuoka Acta Med. 62: 117. (Jap.) (Cited in Kuratsune et al., 1972)

Yamashita, F. 1977. Clinical features of polychlorbiphenyls (PCB)-induced fetopathy. Paediatrician. 6: 20.

Yobs, A.R. 1972. Levels of polychlorinated biphenyls in adipose tissue of the general population of the nation. Environ. Health Perspect. 1: 79.

Yoshimura, H. and H.-A. Yamamoto. 1975. A novel route of excretion of 2,4,3',4'-tetrachlorobiphenyl in rats. Bull. Environ. Contam. Toxicol. 13: 681-688.

Yoshimura, H. and S. Yoshihara. 1976. Toxicological aspects. II. The metabolic fate of PCBs and their toxicological evaluation. In: PCB Poisoning and Pollution, K. Higuchi, Ed. Tokyo, Kodansha Ltd. p. 41-67.

Yoshimura H., S. Yoshihara, N. Ozawa and M. Miki. 1979. Possible correlation between induction modes of hepatic enzymes by PCBs and their toxicity in rats. Ann. NY Acad. Sci. 320: 179-192.

Yoshimura, T. 1971. A case control study on growth of school children with Yusho. Fukuoka Acta Med. 62: 109. (Jap.) (Cited in Kuratsune et al., 1972)

Zauder, H.L. 1979. A possible interaction of PCB and halomethane in man. Anesthesiology. 51: 95.

Zinkl, J.G. 1977. Skin and liver lesions in rats fed a polychlorinated biphenyl mixture. Arch. Environ. Contam. Toxicol. 5(2): 217-228.